ARCHIVES of INTERNAL MEDICINE

VOLUME 76 NOVEMBER-DECEMBER 1945 NUMBER 5

COPYRIGHT, 1946, BY THE AMERICAN MEDICAL ASSOCIATION

ACUTE MYOCARDITIS IN MUMPS (EPIDEMIC PAROTITIS)

COMMANDER DAVID H. ROSENBERG (MC), U.S.N.R.

Mumps (epidemic parotitis) is generally regarded today as an acute systemic disease of virus origin, having a special predilection for the parotid glands. Among the complications most familiar to the physician are acute orchitis, pancreatitis, meningitis and meningoencephalitis. Cardiac involvement has been considered rare and when observed has been described as either acute pericarditis or acute endocarditis. That the virus of mumps may also produce myocarditis was originally suspected by Pujol [1] and by Barbato [2] from their clinical observations, but no satisfactory evidence to support their contention was presented. Later, Manca [3] reported the histologic changes in an instance of acute interstitial fibrinous myocarditis which occurred in a fatal case of mumps. He regarded this as a characteristic reaction to the mumps virus.

In January 1943, I had the unique opportunity of observing an instance of complete heart block in a patient convalescing from epidemic parotitis. From the electrocardiographic changes a diagnosis of acute myocarditis was made, and, in the absence of any other etiologic agent, the disease was ascribed to the virus of mumps. The following report is a brief summary of the case:

CASE 1.—A. C. S., a white sailor aged 20 years, was admitted to the hospital on Jan. 18, 1943, complaining of pain in and swelling of the right side of the face for two days. The past history was noncontributory. Physical examination revealed swelling and tenderness of the right parotid gland, with inflammation of the orifice of Stensen's duct. No other abnormal conditions were observed.

The patient's course was uneventful until the fifteenth day of illness, when he complained of constant retrosternal pain and of dizziness on arising. Examination at that time revealed no abnormalities save for a pulse rate of 35 per minute. The temperature and the respiratory rate were normal. The blood pressure was 110 systolic and 80 diastolic. The electrocardiogram taken on the same day showed evidence of complete auriculoventricular dissociation (fig. 1 *A*). The auricular rate was 104 and the ventricular rate 35 per minute. The P waves were not fixed in the ventricular cycle; they were slightly notched in lead II and inverted in lead CF_4. The T waves were all upright. The blood count was normal except for a leukocyte count of 12,800 per cubic millimeter.

The sedimentation rate was 22 mm. in one hour (Cutler method). Urinalysis showed nothing abnormal. On the following day the ventricular rate was 62 per minute and the rhythm was irregular, with frequent dropped beats. The electrocardiogram (fig. 1 *B*) disclosed second degree auriculoventricular block with the Wenckebach phenomenon and dropped beats. The auricular rate was 70 and the ventricular rate 62 per minute. The P-R interval ranged from 0.22 to 0.44 second. The symptoms gradually subsided in three days, but a harsh systolic murmur became audible at the mitral area. Eight days after the onset of the angina, the electrocardiogram (fig. 1 *C*) was within normal limits, the P-R interval being 0.16 second. The P waves remained notched in lead II and tiny and inverted in lead CF_4. The sedimentation rate and the leukocyte count returned to normal. The systolic murmur diminished in intensity, and at the time that patient was discharged from the hospital it was soft and heard only at the apex when the patient was in the recumbent position.

Early in 1944, another patient was observed who during the course of mumps manifested electrocardiographic evidence of partial heart block which progressed to complete heart block and ultimately returned to normal. Again, the diagnosis of acute myocarditis secondary to the virus of mumps was made. Following is a résumé of this case.

CASE 2.—J. B. B., a white sailor aged 20 years, was admitted to the hospital on Feb. 21, 1944, complaining of pain in and swelling of the right side of the face for two days. The past history was noncontributory. Physical examination revealed swelling and tenderness of the right parotid gland. No other abnormal findings were elicited.

His course was uneventful until the seventh day of illness, when palpitation and precordial pain developed. Examination disclosed an apical rate of 28 per minute. No other abnormalities were noted. The temperature was 101 F. (rectal) and the respiratory rate 16 per minute. The blood pressure was 110 systolic and 52 diastolic. The electrocardiogram taken on the same day showed second degree auriculoventricular block (3:1), with only one out of three auricular impulses being conducted to the ventricles (fig. 2 *A*). The auricular rate was 99 and the ventricular rate 33 per minute. The P waves were fixed in the ventricular cycle. The P-R interval of the conducted sinus impulses was 0.28

This article has been released for publication by the Division of Publications of the Bureau of Medicine and Surgery of the United States Navy. The opinions and views set forth in this article are those of the writer and are not to be considered as reflecting the policies of the Navy Department.

1. Pujol, M.: Oreillons et myocardite, Arch. de méd. et pharm. mil. **69**:527, 1918.

2. Barbato, M.: Un caso di parotite epidemica con complicazioni viscerali multiple, ed esito letale, Riforma med. **41**:1109 (Nov. 23) 1925.

3. Manca, C.: Miocardite da parotite epidemica, Arch. ital. di anat. e istol. pat. **3**:707 (July-Aug.) 1932.

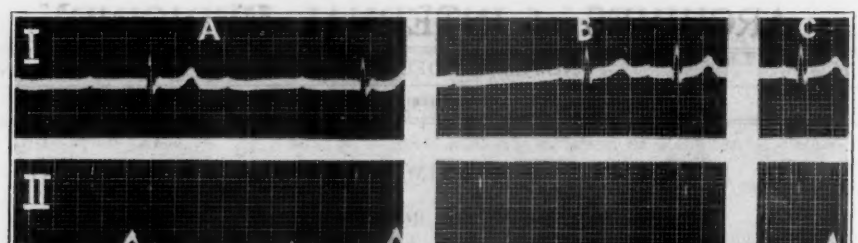


Fig. 1.—Electrocardiograms taken on A. C. S. (case 1) on the fifteenth (*A*), sixteenth (*B*) and twenty-second (*C*) days of illness. *A* shows the characteristic findings of complete auriculoventricular dissociation. In *B*, evidence of improvement is demonstrable in the finding of second degree auriculoventricular block with dropped beats and the Wenckebach phenomenon. In C, the electrocardiogram has returned to normal (see text for details).

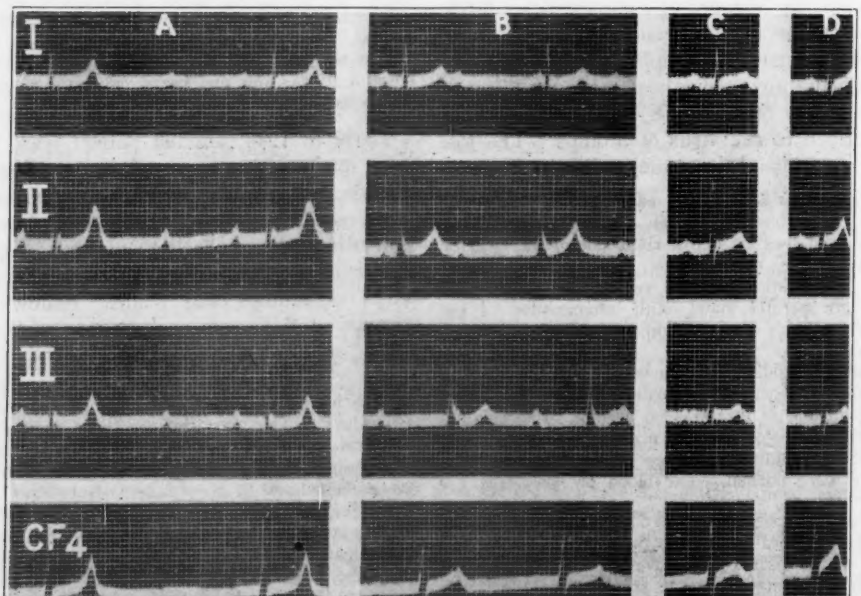


Fig. 2.—Electrocardiograms taken on J. B. B. (case 2) on the seventh (*A*), tenth (*B*), twenty-first (*C*) and eighty-first (*D*) days of illness. In *A*, the electrocardiogram discloses second degree auriculoventricular block (3:1) with dropped beats. The P-R interval of the conducted sinus impulses is 0.28 second. In *B*, the block has increased to complete auriculoventricular dissociation. *C* shows only first degree auriculoventricular block, with prolongation of the P-R interval to 0.23 second, indicating considerable improvement. In *D*, the electrocardiogram has reverted to normal (see text for details).

second. The P waves were peaked in lead II and small and inverted in lead CF_4. The T waves were tall, notched and varied in contour because of superimposed P waves. The sedimentation rate was 33 mm. in one hour. The blood count was normal except for a leukocyte count of 11,400 per cubic millimeter. Urinalysis revealed nothing abnormal. Examination three days later revealed variations in the intensity and quality of the heart tones, suggesting complete heart block. The electrocardiogram on that day showed complete auriculoventricular block, with an auricular rate of 91 and a ventricular rate of 52 per minute (fig. 2 *B*). There was no fixed relation between the P waves and the QRST complexes. The P waves varied in contour (wandering pacemaker). The temperature remained elevated for five days, and palpitation continued for sixteen days. The electrocardiogram taken on the fifteenth day of this complication (fig. 2 *C*) revealed only first degree auriculoventricular block, with a P-R interval of 0.23 second (normal, 0.12 to 0.21 second). All auricular beats were conducted, and the auricular and ventricular rates were 68 per minute. These electrocardiographic abnormalities remained until the seventy-seventh day of the complication, when the electrocardiogram was normal (fig. 2 *D*). The patient was discharged from the hospital six weeks later feeling well. At no time were any cardiac murmurs detectable.

A review of the literature revealed no references to any similar observations. Hence, a study of the electrocardiographic findings in epidemic parotitis was undertaken as an approach to the determination of the incidence, course and prognostic significance of acute myocarditis due to the virus of mumps.

MATERIAL AND METHODS

Electrocardiographic tracings were taken on a consecutive series of 104 adults with mumps who were admitted to a large naval hospital during the height of an epidemic early in 1944. All except 2 patients were men. The ages ranged from 17 to 40 years, the average for the group being 21 years. Standard limb leads and chest lead CF_4 were used in all cases. For 46 of these patients, serial electrocardiograms were made at intervals of two to four days. This study was correlated with determinations of the sedimentation rate (Cutler method) [4] and with blood counts. In addition, a careful search was made for clinical evidence of cardiac involvement. The patients showing electrocardiographic abnormalities were studied further to exclude the possibility of coexistent etiologic factors.

RESULTS

In 16 of the 104 patients studied, significant electrocardiographic evidence of myocardial involvement was found (15.4 per cent). In these no other concomitant disease or infection was recognizable. Electrocardiographic abnormalities were observed in 3 other patients of this series, but in 1 of these the electrocardiographic changes were characteristic of the Wolff-Parkinson-White syndrome, currently believed to be a congenital anomaly; in another, the abnormality was constant and could be attributed to a congenital deformity of the thoracic wall; in the third patient, there was a history of migratory pains in the joints for four months prior to the onset of mumps and physical signs of mitral stenosis, thus indicating the likelihood of rheumatic carditis. These 3 cases have therefore been excluded from consideration.

An analysis of the electrocardiographic abnormalities [5] in the group of 16 patients revealed the following facts [6]: In 4 patients the P waves became diphasic or inverted in one or more leads (fig. 3). Prolongation of the P-R interval was found in 2 instances (fig. 4). In 1 person, the QRS complex in CF_4 became inverted, and in another the voltage in the limb leads decreased. Two patients showed significant elevation of the S-T segments in CF_4, and in 1 of these the S-T segment was bowed upward. In 1 subject, the S-T segment became depressed beyond normal in leads II and III. Changes in the T waves were observed in all the 16 patients, and in 2 of these they were noted in two or more leads (figs. 4 and 5). The T waves became tiny, diphasic or inverted, and by means of serial electrocardiographic studies the evolutionary stages in the development of T wave inversion could be demonstrated. Alterations of the T wave occurred with essentially the same frequency in each of the four leads. In 14 out of the 16 patients, more than one abnormality was found. In the other 2, significant changes in the T wave were found in either lead I or lead II. No specific electrocardiographic pattern was recognizable in this study.

It is noteworthy that in all but 1 patient the electrocardiographic abnormalities were observed between the fifth and the tenth day of illness, and in the majority of these they were present on the eighth or the ninth day. In the other subject, an abnormal tracing was not found until the twentieth day of illness. In 14 patients, the electrocardiographic changes returned to normal in two to thirty-five days. In most of these instances, however, the curves were normal in four to eight days. In the remaining 2 patients, although electrocardiographic evidence of progressive improvement was observed early in the course of this complication, minor residual

4. Cutler, J.: A Graphic Presentation of Blood Sedimentation Test: A Study in Pulmonary Tuberculosis, Am. J. M. Sc. **171**:882 (June) 1926.

5. Rosenberg, D. H.: The Electrocardiographic Changes in Epidemic Parotitis (Mumps), Proc. Soc. Exper. Biol. & Med. **58**:9 (Jan.) 1945.

6. Dr. Louis N. Katz, Director of Cardiovascular Research, Michael Reese Hospital, Chicago, reviewed the electrocardiographic tracings and concurred in the findings and their interpretation.

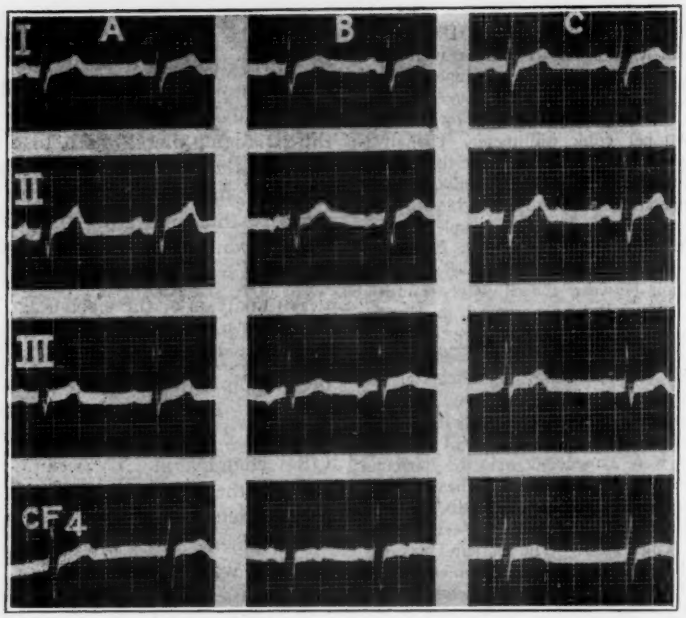


Fig. 3.—Electrocardiograms taken on R. J. P. on the fifth (*A*), ninth (*B*) and thirteenth (*C*) days of illness, demonstrating changes in the P waves and the T waves. In *A*, the electrocardiogram is normal. In *B*, the P waves have become diphasic in lead II, and sharply inverted in lead III. The T waves are of lower amplitude in leads I, II and III and are inverted in lead CF_4. In *C*, the electrocardiogram is again normal.

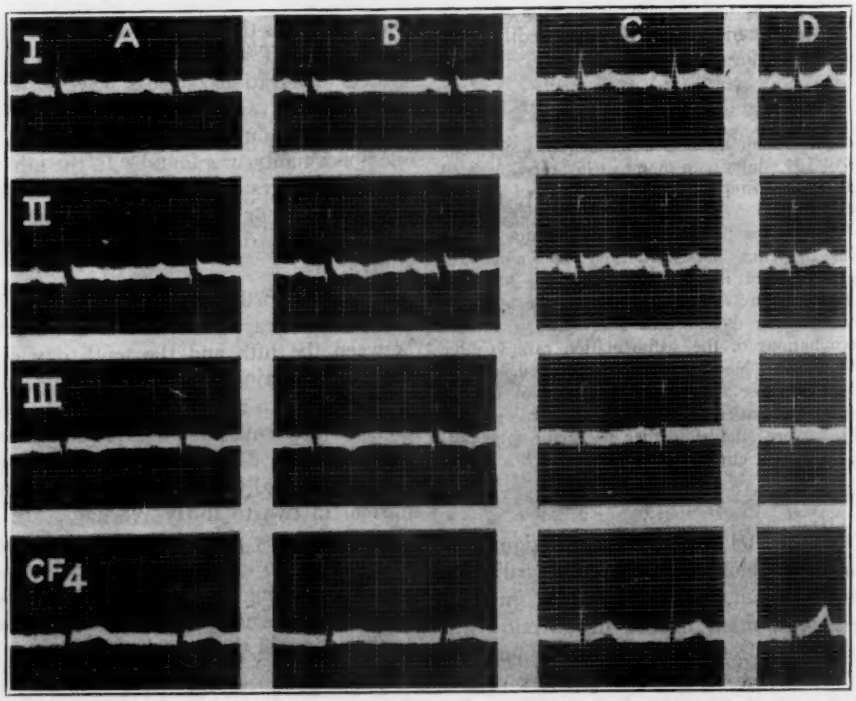


Fig. 4.—Electrocardiograms taken on J. L. C. (case 47) on the seventh (*A*), fourteenth (*B*), thirty-seventh (*C*) and ninety-third (*D*) days of illness, showing prolongation of the P-R interval and T wave changes. In *A*, the P-R interval is prolonged to 0.24 to 0.27 second; the T waves are of low amplitude in lead I and notched in lead II. In *B*, the P-R interval is 0.22 second; the T waves have become inverted in leads I and II and tiny in lead CF_4. In *C*, the P-R interval is 0.22 second; the T waves are now upright in leads I and II and taller in lead CF_4. In *D*, the electrocardiogram has returned to normal.

changes persisted for three months and five months respectively, ultimately reverting to normal.

Precordial pain, either alone or with dyspnea and palpitation, developed in 4 of the 16 patients. In 1 of these, dull precordial pain appeared on the third day of illness and recurred for sixteen days. A soft blowing systolic murmur became audible at the mitral area in all positions and persisted for two months. Two days after the onset of precordial pain, the P waves were inverted in leads II and III, the S-T segment in lead CF_4 was elevated and the T waves in lead III were diphasic. Within thirty-five days the P and T deflections became upright and the S-T segment was normal. In another subject, constricting precordial pain radiating to the left shoulder and arm was noted on the second day of illness. It was accompanied with dyspnea and palpitation and lasted for nine days. Seven days after the onset of these symptoms the T wave in lead I became diphasic and small and returned to normal four days later. In the other 2 patients (cases 47 and 72) mild recurrent precordial pain, palpitation and fatigue developed three months and six weeks respectively after the onset of parotitis. However, the electrocardiograms were abnormal on the seventh and ninth days of illness respectively. In patient 47, the P-R interval was prolonged and the T waves became inverted in leads I and II (fig. 4). In patient 72, the T waves became tiny in lead I and inverted in leads II, III and CF_4 (fig. 5); a soft blowing systolic murmur was audible at the mitral area on the twenty-first day of illness and did not disappear until two months later. When the electrocardiographic changes in these 2 patients were correlated with the time of onset of the cardiac symptoms, it was disclosed that normal electrocardiographic findings had been present for only one to two weeks prior to the resumption of activity in the ward. Longer periods of rest and decreased activity were considered advisable and were followed by a gradual abatement of symptoms. None of the other patients in this study revealed either subjective or objective evidence of any cardiac abnormality except for the aforementioned patient with rheumatic mitral stenosis.

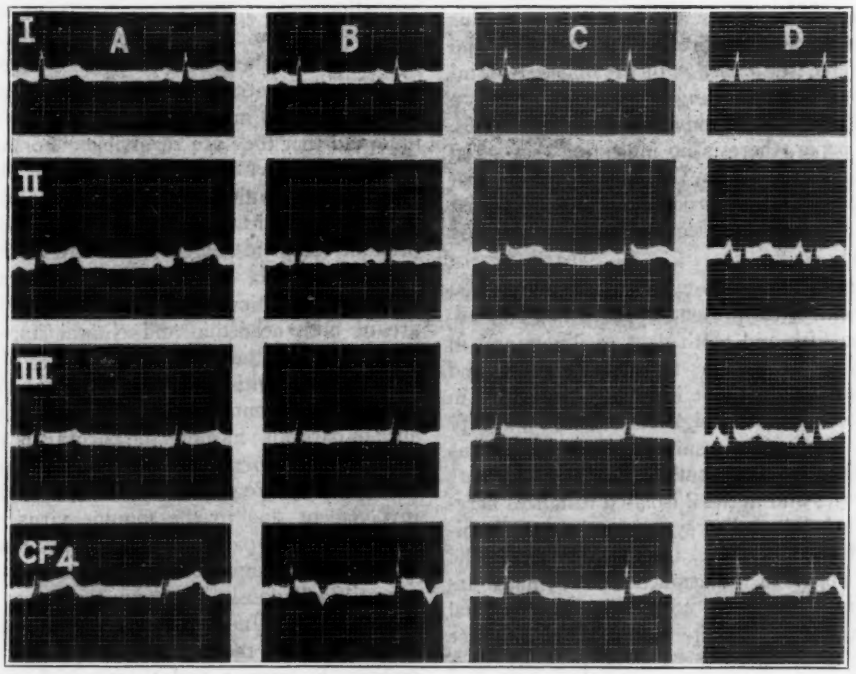


Fig. 5.—Electrocardiograms taken on J. T. A. (case 72) on the fifth (*A*), ninth (*B*), thirteenth (*C*) and thirty-second (*D*) days of illness, demonstrating changes in the T waves. In *A*, the electrocardiogram is essentially normal. In *B*, the T waves have become tiny in lead I and inverted in leads II, III and CF_4. In *C*, the T waves are upright in leads II, III and CF_4 but are of low amplitude in leads I, II and III. In *D*, the electrocardiogram is again normal except for low amplitude T waves in lead I.

The sedimentation rate was elevated in only 5 of the 16 patients, the maximum reading being 22 mm. in one hour. A slight leukocytosis (11,000 to 11,600 leukocytes per cubic millimeter)

was found in 3 patients, and in another the leukocyte count was 14,000 per cubic millimeter. In 7 of the 16 patients acute orchitis developed. It was in 4 of these patients that the leukocytosis appeared and in 3 the sedimentation rate was elevated.

COMMENT

The electrocardiographic conditions observed in 16 of the 104 patients studied would seem to indicate that, contrary to the prevailing literature, myocardial involvement is not a rare event during the course of epidemic parotitis in adults. It should be emphasized that this study was conducted during the peak of an epidemic and that the incidence may be lower during the declining period or in sporadic cases, when the infection is less virulent. Since the completion of this work, attention was directed to Wendkos and Noll's [7] report of a case with prolonged P-R interval and T wave changes occurring among a group of 15 soldiers convalescing from mumps. In their paper, there is no indication that serial electrocardiograms were taken for the 14 patients with normal electrocardiograms or in what stage of recovery the electrocardiographic study was made.

Electrocardiographic evidence of myocardial involvement, when present, appears as a rule between the fifth and tenth days of illness and in this respect is similar to acute orchitis and meningoencephalitis. It is usually transitory in nature, lasting four to eight days in the majority of cases, although occasionally remaining for as long as three to five months. In only 4 patients of this series and in the 2 isolated instances herein recorded did cardiac symptoms become manifest, and in 3 of these a soft blowing systolic murmur appeared. Inasmuch as these murmurs disappeared, they were considered as functional in origin, and no definite diagnostic significance could be attached to them. The presence of a slow or irregular pulse was not necessarily indicative of myocardial involvement, for in a number of such instances the electrocardiograms showed no deviations from normal. It follows, therefore, that the recognition of this complication resides chiefly, if not exclusively, in the results of an electrocardiographic study and that in the majority of cases the cardiac lesion remains at a subclinical level. No sequelae were observed in this group of patients, although convalescence was protracted in 2 instances. Pujol,[1] however, reported 3 soldiers with dyspnea, palpitation, tachycardia and/or precordial pain following an attack of mumps, and I have been informed [8] of other instances in adults with similar symptoms, but without pain, which persisted for several weeks. It is likely that at least in some of my patients the good results may be attributed to the regimen of periodic rest and decreased activity, which was observed for two to three weeks after the electrocardiograms became normal.

The nature of the histopathologic changes in the myocardium is conjectural. Whether or not they represent an acute interstitial fibrinous myocarditis with infiltration of polymorphonuclear leukocytes, as in Manca's case, must be determined by further study. The likelihood of pursuing this problem in human subjects is remote, inasmuch as epidemic mumps is rarely fatal. Information may, however, evolve from observations on experimental animals. From the clinical course and the character of the electrocardiographic observations, the assumption may be drawn that the myocardial changes are usually mild and that they are reversible. For an occasional patient with more extensive involvement, such as those with heart block, failure to recognize this complication may jeopardize the prospects of complete recovery or delay restitution.

The results of this study are provocative of certain implications and considerations which are of both academic and clinical importance. There are authenticated instances of acute orchitis, meningitis and meningoencephalitis due to the virus of mumps which antedate the appearance of parotitis; in some, the parotitis is so slight as to escape recognition. So, too, is it possible to find electrocardiographic evidence of myocardial involvement due to the mumps virus without clinical signs of parotitis, and in such cases, particularly if accompanied with arthralgia, the presumptive diagnosis of rheumatic fever would seem tenable. Indeed, there is a striking similarity between the electrocardiographic abnormalities in mumps and those which were observed [9] in a large series of patients at a naval hospital. It is pertinent to add that other acute infections which are prevalent in epidemic form when the incidence of rheumatic fever is at its peak, such as pneumonia, diphtheria and German measles, were also found with electrocardiographic abnormalities, including prolonged P-R interval. These observations considered collectively cast further doubt on the specificity of the delayed conduction time and suggest that perhaps too much emphasis is being attached to the significance of this finding in the differential diag-

7. Wendkos, M. H., and Noll, J., Jr.: Myocarditis Caused by Epidemic Parotitis, Am. Heart J. 27:414 (March) 1944.

8. Personal communication to the author.

9. Rosenberg, D. H.: Rheumatic Fever in the Adult, paper read at the First Annual Clinical Conference of the Chicago Medical Society, March 1944.

nosis of rheumatic fever. There are doubtless many cases both in the armed forces and among civilians which are labeled as instances of rheumatic fever without sufficient justification other than the presence of arthralgia, unexplained fever or prolonged P-R interval. In view of the growing interest in rheumatic fever and the institution of programs of study dealing with its incidence, prevention and care, there is an urgent need for a more precise definition of rheumatic fever as well as for more specific diagnostic criteria.

It may be argued that perhaps streptococcic infections of the upper respiratory tract (subclinical) or rheumatic fever coexisted in my patients with mumps and that they were responsible for the myocardial changes. Obviously, one cannot settle this point conclusively. It should be pointed out, however, that except for case 1 all the patients with electrocardiographic abnormalities had been participating in the program of continuous mass prophylaxis (sulfadiazine, 1 Gm. daily) then in effect at a large naval training center and that the incidence of infections of the respiratory tract and of rheumatic fever was at the lowest level at the time of this study.

Finally, the observations recorded in this paper present further evidence to substantiate the belief that epidemic parotitis is a generalized infection and that any acute infectious disease, bacterial or viral in origin, is capable of affecting the heart, however infrequent or minimal this may be. As a corollary to this, the cardiac factor may contribute to the causation of some of the symptoms frequently observed during convalescence.

SUMMARY AND CONCLUSIONS

Two isolated cases of mumps complicated by acute myocarditis with electrocardiographic evidence of complete heart block are reported.

In a subsequent study of 104 consecutive cases of mumps in adults, electrocardiographic evidence of myocardial involvement was found in 16 instances (15.4 per cent). The variations in the electrocardiographic observations are presented.

Mumps myocarditis appears as a rule between the fifth and tenth days of illness and is usually mild and transitory. In the majority of instances, it follows a subclinical course and is recognizable chiefly, if not exclusively, by electrocardiographic studies.

Precordial pain, dyspnea and/or palpitation appeared in 4 cases of this series and in the 2 isolated cases.

The implications and the importance of these observations are discussed, particularly as they pertain to the diagnosis of rheumatic fever.

5346 Cornell Ave., Chicago.

PNEUMOTHORAX IN YOUNG ADULT MALES

DESCRIPTIVE STATISTICS IN ONE HUNDRED AND TWENTY-SIX CASES

LIEUTENANT COLONEL JOHN E. LEACH
MEDICAL CORPS, ARMY OF THE UNITED STATES

Spontaneous pneumothorax has been the subject of numerous reports in the literature. With relatively few exceptions,[1] most reports have been concerned with small groups of cases or with isolated patients who have presented unusual features or complications. The reports of larger numbers of cases have usually been drawn from clinics or sanatoriums dealing with tuberculous patients. Reports of this type necessarily must be colored by the relation of pulmonary tuberculosis and spontaneous pneumothorax; consequently, the results are biased in one direction.

The following report is based on a study of 126 patients with 129 episodes of spontaneous pneumothorax observed among military personnel in the Army Air Forces Training Command. Since all military personnel have a roentgenogram of the chest made prior to entrance into the service and since many have roentgenograms made after entering on active duty, the results observed in this group are biased in the opposite direction, namely, away from tuberculosis.

Source of Material for Study.—Certain hospitals in the Command were requested to furnish clinical records and pertinent roentgenograms from all cases of spontaneous pneumothorax in their files. The material was assembled and studied in the Office of the Surgeon of the Army Air Forces Training Command.

The 126 patients reported on by no means represent all the cases of pneumothorax that have been observed. Some records could not be used because of incompleteness, or the complete records had been forwarded to Veterans' facilities or to general hospitals at the time the patients were transferred. However, no other selective principle was used in settling on the population of the 126 cases included in this study.

Results.—So far as possible the data have been assembled and presented in the form of tabulations, both for ease of discussion and reference and for use in comparing or combining with other data that may be reported by others. In connection with each tabulation is presented a comment on the significance of pertinent features or unusual aspects of the data.

General Considerations. — There were 126 male patients with 129 episodes of pneumothorax (1 patient had 1 recurrence and 1 patient had 2 recurrences during the period covered by this survey). There were 5 ground officers, 80 nonflying enlisted men, 4 pilots and 1 bombardier, 1 navigator, 3 aerial gunners, 2 student flying officers and 30 aviation cadets. Ages ranged from 18 to 41 years, averaged 24.8 years, with a standard deviation of 5.6 years. There were no immediate fatalities. The number of fatalities among those transferred (10 patients) beyond the jurisdiction of this command is unknown.

Special Considerations.—It has been taught that the onset of spontaneous pneumothorax is usually sudden and is associated with rather pronounced symptoms of dyspnea, pain in the chest, pallor and evidences of cardiovascular embarrassment even to the point of shock. In an appreciable portion of these patients the onset was gradual (table 1). In another appreciable portion the initial symptoms were either nonexistent (ranging from vague discomfort in the chest to absolutely no symptoms) or mild in character. The difference between those exhibiting no symptoms or only mild ones (63.5 per cent) and those exhibiting moderate and severe symptoms (36.5 per cent) is statistically significant.

1. (*a*) Ornstein, G. G., and Lercher, L.: Spontaneous Pneumothorax in Apparently Healthy Individuals, Quart. Bull. Sea View Hosp. **7**:149-187 (April) 1942. (*b*) Kjaergaard, H.: Spontaneous Pneumothorax in Apparently Healthy, Acta med. Scandinav., 1932, supp. 43, pp. 1-159 and 1-93; cited by Blackford, S. D.: Spontaneous Pneumothorax in College Students, J. A. M. A. **113**:737-739 (Aug. 26) 1939. (*c*) Olbrechts, E.: Le pneumothorax spontané "idiopathique" Bénin, Ann. de méd. **27**:429-483 (May) 1930. (*d*) Norris, J. L.: Spontaneous Pneumothorax; Industrial Experience with 25 Cases, New York State J. Med. **40**:504-506 (April 1) 1940. (*e*) Bassel, P. M.: Spontaneous Pneumothorax, Texas State J. Med. **32**:696-700 (Feb.) 1937. (*f*) Kirshner, J. J.: Spontaneous Pneumothorax, Am. J. M. Sc. **196**:704-709 (Nov.) 1938. (*g*) Blackford, D.: Spontaneous Pneumothorax in College Students, J. A. M. A. **113**:737-739 (Aug. 26) 1939.

It is possible that the generally high state of physical fitness of the men contributed to the mildness of the symptoms. There was no association between the severity of the reported symptoms at the onset of the attack and the degree of pulmonary collapse.

TABLE 1.—*Onset and Initial Symptoms of the Attack (One Hundred and Twenty-Nine Attacks)*

	Number	Per Cent of Number	Per Cent of Number with Unknown Onset Eliminated
Onset			
Gradual	23	17.8	20.2
Sudden	90	69.8	79.8
Unknown or unstated	16	12.4	
Total	129	100.0	100.0
Initial symptoms	**Number**	**Per Cent of Number**	**Per Cent of Number with Unstated Symptoms Eliminated**
None	8	6.2	6.8
Mild	67	52.0	56.7
Moderate	6	4.7	5.1
Severe	37	28.6	31.4
Unstated	11	8.5	
Total	129	100.0	100.0

In table 2 it is significant to note that in 76 patients there was nothing in the past medical history that had any reasonable association with the onset of spontaneous pneumothorax. In addition, 18 patients had infections of the upper respiratory tract, many of which were

TABLE 2.—*Significant Past Medical History up to Onset (One Hundred and Twenty-Nine Attacks)*

	Number	Per Cent
Noncontributory (4 found on routine roentgenographic examination)	76	59.0
Concomitant infection of the upper respiratory tract (5 without cough; 13 with cough)	18	14.0
History of previous pleural pain	6	4.6
Concomitant acute pleuritis	3	2.3
Previous empyema	1	0.8
Chronic pulmonary emphysema	1	0.8
Concomitant pneumonia (1 lobar; 1 bronchopneumonia; 1 virus)	3	2.3
Exposed to or studied for tuberculosis	5	3.9
Inactive pulmonary tuberculosis	1	0.8
Indirect trauma (struck on face)	1	0.8
Possible previous pneumonia	3	2.3
Previous pneumothorax	8	6.2
Possibly associated with "flight" in low pressure chamber	3	2.3
Total	129	100.0

mild. Although 13 had an infection of the upper respiratory tract associated with cough, in only 1 instance did the pneumothorax occur during a period of coughing.

Those patients who had a history of previous pneumothorax will be commented on separately, since they are of themselves an interesting group. The remaining categories are too scattered and do not constitute sufficiently large groups for separate consideration.

Three patients, 2 of whom gave vague histories dating from a low pressure chamber "flight" one month prior to the onset of the pneumothorax and 1 in whom the onset of pneumothorax occurred during a "flight," are included only to make a point. When one considers the large number of "man flights" made daily in low pressure chambers in this Command entirely unassociated with pneumothorax, these 3 instances seem extraordinarily unimportant.

None of these 129 onsets occurred during aerial flight.

There have been many discussions in the past concerning the relation between physical activity and the production of pneumothorax. A division of the cases into groups according to amount of physical activity (none, mild or moderate activity and extreme activity), based on the premise that with chance factors alone operating there would be an equal distribu-

TABLE 3.—*Physical Activity at Onset*

	Number	Per Cent of Number	Per Cent of Number with Unknown or Unstated Activity Eliminated
At rest (30 in bed; 20 sitting)	50	38.8	48.6
Mild physical activity (6 leaning over; 11 standing; 20 walking; 2 climbing stairs)	39	30.2	37.8
Extreme physical activity (4 running; 8 violently exercising; 1 coughing; 1 fighting)	14	10.8	13.6
Unknown or unstated (includes 4 discovered in routine roentgenographic examination)	26	20.2	
Total	129	100.0	100.0

tion in the three categories, brings out two important features (table 3). In a significantly higher percentage (48.6 per cent) of cases than would be grouped together by chance, the onset of pneumothorax was associated with a minimum of physical activity. In a significantly lower percentage (13.6 per cent) of cases, the onset was associated with extreme physical activity.

This distribution suggests two explanations: First, the amount of physical activity has little to do with the onset of the average case of pneumothorax or, second, the cause of pneumothorax occurring at rest is quite different from the cause of that occurring during exercise.

There was no association between the amount of physical activity and either the symptoms or the degree of collapse at the onset.

It can be seen by a glance at table 4 that the valvular type of pneumothorax was an unusual occurrence in this group.

The more frequent occurrence of pneumothorax on the right side has been noted before.[1a] In this group 59.8 per cent of the occurrences were on the right side (table 4). This difference from a chance distribution is significant at the 1 or 2 per cent level. When this group is combined with the cases reported by Ornstein and Lercher [1a] (25 single attacks on the right side and 16 on the left) the difference between the frequency of right-sided and left-sided pneumothorax is highly significant (0.1 per cent level).

It is not apparent from these data why spontaneous pneumothorax occurs more frequently on the right side.

TABLE 4.—*Type and Side of Occurrence of Pneumothorax*

	Number	Per Cent of Number	Per Cent of Number with Unstated Side Eliminated
Type of pneumothorax			
Closed	127	....	98.5
Valvular	2	....	1.5
Side of pneumothorax			
Right	76	58.9	59.8
Left	50	38.8	39.4
Bilateral	1	0.7	0.8
Unstated	2	1.6	

TABLE 5.—*Degree of Collapse and Rate of Reexpansion*

Days to Complete Reexpansion	Group I Up to 50% Collapse, Number	Group II Greater Than 50% Collapse, Number	Both Groups, Number
0 to 10	6	1	7
11 to 20	32	1	33
21 to 30	12	4	16
31 to 40	4	12	16
41 to 50	1	5	6
51 to 60	0	8	8
61 to 70	1	1	2
71 to 80	0	2	2
81 to 90	0	0	0
91 to 100	0	3	3
101 to 110	0	0	0
111 to 120	0	1	1
Total	56 (59.5%)	38 (40.5%)	94 (100%)*
Mean in days †	19.43	48.39	31.14
Standard deviation of distribution	10.29	23.16	21.96
Standard error of mean	1.39	3.71	2.27

* For 32 patients, not all the roentgenograms were submitted. Hence, these cases could not be included.
† The critical ratio of difference between means of groups I and II is 7.3.

The estimation of the per cent of collapse in pneumothorax is variable because of differences of method among roentgenologists and variations in technic in making roentgenograms. In compiling the data shown in table 5, these differences have been minimized as much as possible by making only two groups, namely, those with pulmonary collapse up to 50 per cent and those with collapse over 50 per cent.

These data show (*a*) a suggestion of a likelihood that the degree of pulmonary collapse will be less than 50 per cent and (*b*) a highly significant difference between the mean number of days required for complete reexpansion to take place in groups I and II. As a matter of fact, in this distribution slightly over one half of all the patients had degrees of pulmonary collapse estimated at below 33 per cent.

The cause of the pneumothorax could not be determined for the great majority of the patients, despite the fact that all the usual diagnostic measures were carried out. It is possible that the condition in a goodly portion of those patients transferred to general hospitals and veterans' facilities was tuberculous in origin (table 6).

TABLE 6.—*Cause (One Hundred and Twenty-Nine Attacks)*

	Number
Undetermined	107
Emphysema	1
Proved pulmonary tuberculosis	1
Pneumonia	1
Pleurisy	1
Postoperative	1
Unknown because of transfer or because still in hospital	17
Total	129

For the sake of emphasis it should be stated that subpleural emphysematous blebs or visceroparietal pleural adhesions could not be demonstrated on the roentgenograms. It is possible, of course, that they might have been demonstrated by views of the thorax from other angles.

From table 7 it is evident that complications in the course of the pneumothorax were infrequent. The average length of hospitalization (39.9 days) was determined largely by the average number of days (31.1 days) required for the air to be reabsorbed, plus additional days (average eight days) for further convalescence.

TABLE 7.—*Complications in the Course of Pneumothorax*

Complications	Number	Per Cent
None	102	79.2
Slow absorption	3	2.3
Thoracentesis for excessive pain or dyspnea	2	1.5
Hemothorax	2	1.5
Increase in degree of pneumothorax	4	3.1
Associated injuries or other diseases	2	1.5
Pneumonia and pleurisy	4	3.1
Unstated or unknown	10	7.8
Total	129	100.0

Days of Hospitalization	
Range	4 to 135 days
Mean days	39.9 days
Standard deviation of distribution	26.6 days

TABLE 8.—*Disposition of Personnel*

	Number	Per Cent
Retained in military service (82 full duty; 11 limited service)	93	73.8
Certificate of disability discharge	13*	10.3
Still in hospital	7	5.5
Unknown because of transfer	10	7.9
Unstated disposition	3	2.4
Total	126	99.9

* One was separated because of constitutional psychopathy and 1 because of nonspecific ulcerative colitis.

TABLE 9.—*Disposition of Flying Personnel*

Disposition	Number
Restored to flying after leaving hospital (1 pilot, 1 student officer, 2 aerial gunners, 1 aviation cadet)	5
Temporarily suspended from flying (1 pilot, 1 bombardier, 1 navigator, 1 aerial gunner, 1 student officer, 1 aviation cadet)	6
Grounded for 12 months (aviation cadet)	1
Indefinitely suspended (all aviation cadets)	19
Unstated disposition (1 pilot, 6 aviation cadets)	7
Still in hospital (1 pilot, 2 aviation cadets)	3
Total	41

history of previous pneumothorax (table 10). The 7 patients had a total of 11 previous episodes of pneumothorax. It is interesting that 5 of the 7 patients had recurrences on the right side. It is also noteworthy that the recurrences were not associated with unusual physical activity. There were several additional patients who gave suggestive histories of previous attacks of pain in the chest and/or dyspnea, but these could not be included in this group because of the indefiniteness of the histories. One cannot draw any conclusions from these cases on the degree of tendency of pneumothorax to recur, since the group is too small and the time period covered is too short.

COMMENT AND SUMMARY

As noted before, this group of patients, previously screened for chronic pulmonary disease (chiefly pulmonary tuberculosis) prior to entrance on active duty, cannot be considered a general sample of the adult male population. The screening accomplished by the preservice roentgenogram of the chest largely negates the known relation between pulmonary tuberculosis and pneumothorax. However, by so doing, it brings to the fore those types of pneumothorax which for want of a better term, have been designated "spontaneous pneumothorax in healthy adults," noted by Hall [2] and later by Kjaergaard.[1b] Olbrechts [1c] described a similar type of pneumothorax among French army conscripts, "the pneumothorax of conscripts."

From the results obtained in this group so heavily weighted with the nontuberculous type of

TABLE 10.—*Recurrent Pneumothorax* *

Patient	Age	Number of Previous Attacks	Side of Previous Attack	Side of Present Attack	Degree of Collapse Present Attack (%)†	Physical Activity at Onset of Present Attack	Days to Complete Absorption
1	24	2	? ?	R	+50 ?	Walking	38
2	19	1	L	R	+50	Marching and counting cadence	35
3	19	2	R L	R	?	?	?
4	34	2	R L	L	—50	Sleeping	36
5	21	2	R R	R	?	Marching	?
6	23	1	?	L	+50	Leaning over	43
7	26	1	L	R	—50	Mild physical activity	27
Total 7		11	4L 4R 3 ?	2L 5R			

* All recurrences, except one, were within 1 to 2 years of the present attack.
† The symbol +50 indicates greater than 50 per cent collapse; —50 indicates less than 50 per cent collapse.

Of 126 persons, 93 were retained in the military service (table 8), a salvage of 73.8 per cent. Of the remaining persons, 13 were known to have been separated from the service, and possibly half of the 10 patients who were transferred to other medical facilities (usually because of troublesome complications or for administrative reasons) were also discharged for physical disability.

Of the 41 patients who were flying personnel, the disposition varied considerably (table 9). The trained personnel (pilots, bombardier, navigator and aerial gunners) and the student officers were either restored to flying directly after hospitalization or suspended from flying up to twelve months and then restored. The aviation cadets and aviation students, for the most part, were physically disqualified for further flying duty.

There were 7 patients (1 with 2 recurrences during the period covered) who gave a positive

2. Hall, F. de H.: On Cases of Pneumothorax in Persons Apparently Healthy, Tr. Clin. Soc., London **20**:153-161, 1886-1887; cited by Van Astrand, H. S.: Idiopathic Spontaneous Pneumothorax, Cleveland Clin. Quart. **7**:178-183 (July) 1940.

spontaneous pneumothorax some interesting observations can be made:

1. Not infrequently the onset may be gradual rather than sudden.

2. Initial symptoms are frequently absent (none, vague symptoms or atypical symptoms) or mild in degree.

3. A positive or associative history of previous pulmonary disease is not usually found (an idiopathic pneumothorax).

4. In 85 out of 100 cases the onset of the attack occurred when the patient was at rest (48.6 per cent) or during mild physical activity (37.8 per cent).

5. The side of the pneumothorax is more frequently the right.

6. The closed type of pneumothorax is most common.

7. The degree of collapse is frequently less than one third, and on the average the air can be expected to be absorbed in about twenty days in those who have less than 50 per cent pulmonary collapse.

8. A search for the causative factor in these cases is usually fruitless.

9. Complications are not frequent and not particularly dangerous if treated by standard procedures.

10. Three out of 4 patients were salvaged for further military duty.

11. Except in a long time study of a large number of cases, inferences concerning the degree of tendency for pneumothorax to recur are not valid.

12. The immediate or remote cause of this type of pneumothorax is still unsettled. The theories, spontaneous rupture of a weakened portion of the pleura, tearing of pleural adhesion, rupture of subpleural emphysematous blebs, interstitial emphysema leading to a subpleural collection and finally rupture [3] and wearing through of the pleura through the process of normal denudation,[4] have not been disproved. Conversely, none of these theories can be strongly advocated on the basis of these observations. The theories of a tearing of a pleural adhesion or rupture of an emphysematous bleb are not supported by these cases, since residual pleural adhesions and emphysematous blebs could not be demonstrated on the roentgenograms. However, judging from the work of others, the rupture of an emphysematous bleb still seems the most likely immediate cause. In addition, there was no demonstrable association with pulmonary tuberculosis.

13. So far as these data are concerned, there is no demonstrable relation between flying and spontaneous pneumothorax.

3. Hamman, L.: A Note on the Mechanism of Spontaneous Pneumothorax, Ann. Int. Med. **13**:923-927 (Dec.) 1939.

4. Schomer, A., and Ehrlich, D. E.: M. Bull. Vet. Admin. **11**:206, 1935.

STREPTOMYCIN FOR CERTAIN SYSTEMIC INFECTIONS AND ITS EFFECT ON THE URINARY AND FECAL FLORA

HOBART A. REIMANN, M.D.; ALISON H. PRICE, M.D.
AND
WILLIAM F. ELIAS, PH.D.
PHILADELPHIA

Streptomycin, an antibiotic substance derived from Actinomyces griseus, when injected parenterally appears in the blood, urine and spinal fluid in amounts theoretically bacteriostatic or bactericidal for certain gram-negative bacilli known to be resistant to penicillin and to sulfonamide compounds.[1] When given orally, only traces appear in the blood and urine, since most of it is excreted in the feces, where it exerts a suppressive effect on various bacteria. It is relatively nontoxic. These facts, together with suggestive evidence of the therapeutic value of streptomycin in several cases of typhoid [1a] and in experimental infections in animals,[2] warrants further trial of the drug in several directions; namely, (*a*) for more patients with typhoid; (*b*) for infections caused by gram-negative bacilli, especially brucellosis, bacillary dysentery, tularemia, plague, cholera and Escherichia coli and Hemophilus influenzae infections; (*c*) for infections of the urinary tract, and (*d*) to study its effect on the flora of the intestinal tract.

In this paper, report is made of the effect of streptomycin in 5 more patients with typhoid, 3 with brucellosis, 1 with tuberculous meningitis and 5 with infection of the urinary tract, and in 6 patients to study its effect on the flora in the feces. Including 5 patients in a previous report, streptomycin was given to 35 patients. Report of the use of streptomycin in 10 cases of cholera will appear elsewhere.

Methods—The preparation of solutions of streptomycin hydrochloride and the methods of its intravenous, intramuscular and oral administration were the same as described before.[1a] Assay of the amount of streptomycin in the blood, urine, feces and spinal fluid was made according to Stebbins' method.[3] The potency of the various lots of streptomycin used varied from 130 to 430 units per mg. Amounts of streptomycin recorded in these studies are expressed in terms of weight of pure streptomycin base according to the newly adopted plan whereby 1 microgram represents 1 unit and 1 Gm. represents 1,000,000 units. Estimation of the number of bacteria in the urine and feces was made by standard methods.

REPORT OF CASES

TYPHOID

CASE 1.—H. B., a woman aged 33, was admitted to the Germantown Hospital, service of Dr. Thomas Garret, in a stuporous state with high fever and bacteremia with Eberthella typhosa. Streptomycin, 4 Gm. daily, was given intravenously beginning on the tenth day of the disease. The temperature, which was 105 F., declined during three days of therapy to 102 F. (chart 1), but the stupor deepened into coma and death occurred on the fourteenth day, after about 15 Gm. had

From the Jefferson Medical College and Hospital (Dr. Reimann and Dr. Price) and the Wyeth Institute of Applied Biochemistry (Dr. Elias).

Streptomycin hydrochloride was generously supplied by Merck & Co., Inc., Rahway, N. J. Mrs. Jane Durso assisted with the technical details of the study.

1. (*a*) Reimann, H. A.; Elias, W. F., and Price, A. H.: Streptomycin for Typhoid: A Pharmacologic Study, J. A. M. A. **128**:175-180 (May 19) 1945. (*b*) Elias, W. F., and Durso, J.: Blood, Urine, and Fecal Levels of Streptomycin in the Treatment of Human Infections of E. Typhosa, Science **101**:589-591 (June 8) 1945. (*c*) Hinshaw, H. C., and Feldman, W. H.: Streptomycin in Treatment of Clinical Tuberculosis: A Preliminary Report, Proc. Staff Meet., Mayo Clin. **20**:313-318 (Sept. 5) 1945. (*d*) Anderson, D. G., and Jewell, M.: The Absorption, Excretion and Toxicity of Streptomycin in Man, New England J. Med. **233**: 485-491 (Oct. 25) 1945. (*e*) Zintel, H. A., and others: Studies in Streptomycin in Man: I. Absorption, Distribution, Excretion and Toxicity, Am. J. M. Sc. **210**:421-430 (Oct.) 1945. (*f*) Heilman, D. H., and others: Streptomycin: Absorption, Diffusion, Excretion and Toxicity, ibid. **210**:576-584 (Nov.) 1945, (*g*) Herrell, W. E., and Nichols, D. R.: The Clinical Use of Streptomycin: A Study of Forty-Five Cases, Proc. Staff Meet., Mayo Clin. **20**:449-462 (Nov. 28) 1945.

2. Heilman, F. R.: Streptomycin in the Treatment of Experimental Tularemia, Proc. Staff Meet., Mayo Clin. **19**:553-559 (Nov. 29) 1944. Feldman, W. H., and Hinshaw, H. C.: Effects of Streptomycin on Experimental Tuberculosis in Guinea Pigs, ibid. **19**:593-599 (Dec. 27) 1944. Heilman, F. R.: Streptomycin in the Treatment of Experimental Infections with Micro-Organisms of the Friedländer Group (Klebsiella), ibid. **20**:33-39 (Feb. 7) 1945. Smith, M. I., and McClosky, W. T.: The Chemotherapeutic Action of Streptomycin and Promin in Experimental Tuberculosis, Pub. Health Rep. **60**:1129-1138 (Sept. 28) 1945.

3. Stebbins, R. B., and Robinson, H. J.: A Method for Determination of Streptomycin in Body Fluids, Proc. Soc. Exper. Biol. & Med. **59**:255-257 (June) 1945.

been given. Bacteremia persisted, although the amount of streptomycin in the blood varied from 11 to 22 micrograms per cubic centimeter (22 to 44 micrograms per cubic centimeter of serum), which amount was more than that needed (9 micrograms) to kill the same strain of E typhosa on agar. Her spinal fluid contained 20 micrograms of streptomycin per cubic centimeter, and the usual 30 to 50 per cent of the amount injected was excreted in the urine.

Although this severely sick patient was treated early in the disease with theoretically adequate amounts of streptomycin, the blood was not cleared of E. typhosa and death occurred. The typhoid bacillus from this patient

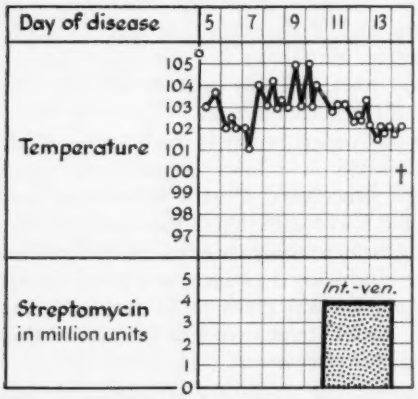


Chart 1 (case 1).—Typhoid. The patient received 4 Gm. (4,000,000 units) of streptomycin intravenously for over three days but died despite the presence of 22 micrograms of streptomycin per cubic centimeter in the blood while only 9 micrograms killed the infecting bacillus in broth culture.

was killed by 9 micrograms of streptomycin per cubic centimeter of agar, as compared with a stock strain, which was killed by 2 micrograms but not by 1 microgram, and with the strain obtained in the epidemic,[1a] which was inhibited by 6 micrograms.

CASE 2.—L. B., aged 21, was admitted to the service of Dr. Sumner Cross at the Abington Memorial Hospital on the fifth day of disease. Although the temperature varied from 103 to 105 F., she never appeared to be toxic and was regarded as having a mild attack, although E. typhosa was cultivated several times from the blood and from the feces. Streptomycin in doses of 4 Gm. daily was given intravenously, beginning on the twelfth day of disease. Within thirty hours the temperature fell to normal and recovery occurred (chart 2). Amounts between 11 and 20 micrograms per cubic centimeter of blood were present during therapy. An amount estimated as between 2 and 5 micrograms appeared in the spinal fluid. Treatment was given in large doses for four days, after which the dose was reduced to 2 Gm. and then to 1 Gm. A total of about 19 Gm. was given.

Recovery seemed to have been brought about by streptomycin in this patient. The infecting bacillus, however, was more resistant than any other strains of E. typhosa tested and withstood 16 micrograms of streptomycin per cubic centimeter but not 18 micrograms when cultured on agar. This quantity was only slightly exceeded by the amounts measured in the blood. Evidence in this case and in case 1 suggests that susceptibility of the infecting strain of E. typhosa to streptomycin and the amounts of drug present in the blood bear no close correlation as regards the clinical outcome. Therapeutic failure in case 1 occurred with a relatively sensitive strain and large amounts of drug in the blood, while recovery occurred in case 2 with the same dosage, but with lesser amounts in the blood and a more resistant strain. Either the inhibition test is not sensitive enough for minor variations in resistance or other obscure factors in the host also play a role. If, according to Goodpasture's view, typhoid bacilli reside and multiply intracellularly in plasma cells and macrophages, they may not be affected by streptomycin unless they escape into the blood or tissues.

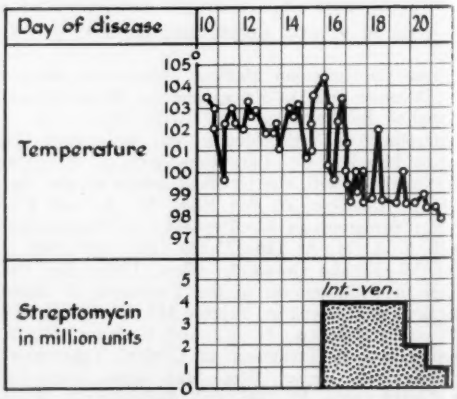


Chart 2 (case 2).—Typhoid. The patient received 4 Gm. (4,000,000 units) of streptomycin intravenously for four days and then smaller amounts. Recovery occurred, but only 11 to 20 micrograms of streptomycin appeared in the blood, while as much as 18 micrograms per cubic centimeter was needed to kill the bacillus in broth culture.

Three more patients, whose records need not be described in detail, were treated. Two, whose temperatures declined slowly over a week and who recovered, received 20 Gm. and 45 Gm. respectively, and 1, who received 24 Gm., died.

BRUCELLOSIS

If streptomycin is bacteriostatic or bactericidal for E. typhosa in the body, it should, theoretically, be even more so for Brucella suis.

since three strains from patients treated were killed on culture with as little as 0.5 microgram per cubic centimeter of agar, as compared with 6 to 18 micrograms needed to kill E. typhosa isolated from patients.

CASE 3.—C. M., a physician aged 41, had an attack of supposed grip lasting two weeks in February. He was then well until about March 1, when malaise, weakness, chilliness and fever (temperature 102 F.) recurred. Later there were severe chills, drenching sweats, insomnia, headache, aching and constipation characteristic of a severe attack. The titer of a brucella agglutinin test was 1:640. Large doses of sulfadiazine and later of penicillin were given in spite of lack of evidence of their value for brucellosis. A course of injections of vaccine was likewise ineffective.

He was admitted to this hospital on the twenty-sixth day of the relapse, which began March 1. The important data are the fever chart (chart 3), leukopenia, agglutinin for brucella in titer of 1:2,000 (had received vaccine) and a blood culture positive for Brucella suis. The growth of this bacterium was inhibited by 0.5 microgram of streptomycin but not by 0.1 microgram per cubic centimeter of agar.

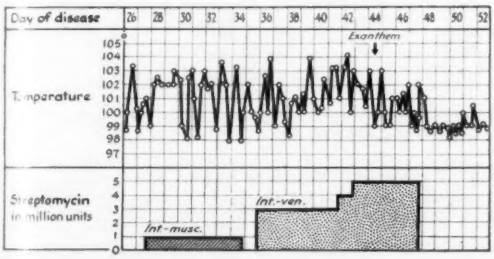

Day of disease
26 28 30 32 34 36 38 40 42 44 46 48 50 52
Exanthem
Temperature
105 104 103 102 101 100 99 98 97
Streptomycin in million units
5 4 3 2 1 0
Int.-ven.
Int.-musc.

Chart 3 (case 3).—Brucellosis. There was no response to small doses of streptomycin intramuscularly or intravenously, but after the forty-second day, when 5 Gm. (5,000,000 units) were given daily, the temperature began to decline. The Br. suis responsible was inhibited by as little as 0.5 micrograms per cubic centimeter of broth; the blood contained as high as 27 micrograms per cubic centimeter.

Because of previous experience with typhoid [18] in which 12 micrograms per cubic centimeter of blood appeared after daily intramuscular injection of 1 Gm., this dosage was first used for seven days. Three cubic centimeters of a solution of streptomycin in distilled water (each cubic centimeter containing about 45,000 micrograms) was given every three hours. Aside from local pain no other untoward symptoms were noted. On this dosage only from 1 to 4 micrograms appeared in the blood, 40 per cent was excreted in the urine and none in the feces. Since no clinical improvement occurred, the dosage was increased to 3 Gm. daily and given intravenously (chart 3). The amount in the blood rose to 9 and 13 micrograms per cubic centimeter of blood, but still there was no clinical improvement after six days. The dose was increased to 4 Gm. and then to 5 Gm., with a rise in the amount in the blood to 27 micrograms per cubic centimeter, which is about sixty times the amount needed to kill the infecting bacilli in vitro. Doses of this size caused about 2,800 micrograms per cubic centimeter to appear in the urine and 11 micrograms per cubic centimeter in the spinal fluid but only traces in the feces. There was no evidence of toxicity other than a diffuse erythematous eruption lasting twelve hours on the forty-fourth day.

After dosage at 5 Gm. was begun, the fever gradually declined for five days and ended abruptly on the forty-eighth day, with coincident clinical improvement. A total of about 54 Gm. of streptomycin was given. The patient returned to his home on April 23, after six days without a rise of temperature over 99 F. During the second week the temperature occasionally rose to 101 F. and once to 102.3 F. In the third week, there were occasional chills with spikes of temperature to 104 F., but the patient felt well enough to get out of bed. Thereafter recovery took place.

In this instance the sensitivity of the infecing Brucella to streptomycin on agar was not a reliable guide to the amount needed in the blood to control the infection. Recovery in this patient was probably spontaneous and not related to therapy, yet the fever declined during treatment after large doses were used. The subsequent recrudescence shows that the infection persisted and suggests the need for continuing treatment for some time after defervescence.

CASE 4.—G. V. B., aged 31, a veterinary assistant, noted slight fever in mid-April, which persisted and increased until he was admitted to Abington Memorial Hospital, May 5, as a patient of Dr. E. S. Vollmer.

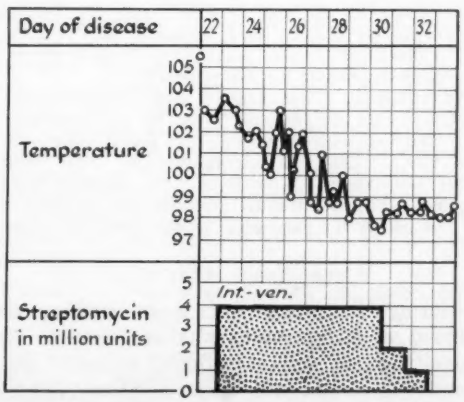


Chart 4 (case 4). — Brucellosis. There was prompt defervescence and recovery during treatment with 4 Gm. (4,000,000 units) of streptomycin intravenously. Br. suis was killed by 0.5 microgram per cubic centimeter of broth; the blood contained 12 to 19 micrograms per cubic centimeter.

Drenching sweats occurred and fever increased to 104 F. Physical examination revealed nothing essentially abnormal except that the patient appeared to be severely sick. The leukocytes numbered 5,000, and 60 per cent were lymphocytes. The serum agglutinated Brucella in a titer of 1:360, and Br. suis grew in a culture of the blood. The patient was transferred to this hospital on May 10, about the twenty-second day of disease.

Because of the experience in case 3, therapy was started with large dosage. Streptomycin was given intravenously in doses of 4 Gm. daily. Shortly thereafter, the temperature began to decline steadily until it reached normal on May 17, after six days of treatment with 24 Gm. The dose was then reduced to 2 Gm., then to 1 Gm. (chart 4). The amount of streptomycin in the blood varied from 12 to 19 micrograms per cubic centimeter, and in the serum from 25 to 35 micrograms. The infecting Brucella, as in case 3, was inhibited by 0.5 microgram of streptomycin per cubic centimeter of agar.

The appearance of the temperature curve (chart 4) and the clinical improvement strongly suggest that streptomycin played a role in the recovery. No relapse has occurred to date.

CASE 5.—A. A., aged 6, was noted by her parents to have fever, gradually rising to 104 F., in February 1945. She recovered after several weeks and was apparently well until April 28, when fever recurred, and on May 5 she entered the Jewish Hospital as a patient of Dr. Schless. Her temperature rose to 104 F. It gradually fell to normal after a week, remained normal for four days and rose again to 105 F. During this time no other physical abnormalities except pallor, sweating and hepatomegaly were noted. The leukocytes numbered 6,000, and 65 per cent of them were lymphocytes. Agglutination for Brucella occurred in a titer of 1: 160. A cutaneous test with brucella vaccine elicited a strongly positive reaction. A diagnosis of brucellosis was made, and the patient was transferred to this hospital for treatment.

At this time, about the nineteenth day of the current bout, the temperature was 105 F., the leukocytes numbered 5,000, 60 per cent of which were lymphocytes, agglutinins for brucella were present in titer of 1: 500 and a blood culture was positive for Brucella abortus. The child weighed 25 Kg. (62 pounds).

Streptomycin, 3 Gm. daily, a large amount in proportion to her weight, was given by the intravenous drip method and continued for about eight days (24 Gm.). Only 12 micrograms per cubic centimeter was present in the blood. No beneficial effect resulted. The Br. abortus involved was inhibited by 0.5 but not by 0.1 microgram of streptomycin in agar.

TUBERCULOUS MENINGITIS

During the course of these studies, the amount of streptomycin was measured in the spinal fluid of several patients, as noted incidentally in certain case reports and listed in table 1. Assuming the meninges to be unaffected in each instance, it is obvious that streptomycin injected parenterally enters the spinal fluid in variable, but in theoretically bactericidal, amounts, as shown in table 1. In case 1 twice the amount appeared with 4 Gm. doses as with 5 Gm. in case 3.

It is questionable whether it is more important to attain a high concentration of the drug in the spinal fluid by intrathecal injection rather than in the meningeal and other tissues by the parenteral route for the treatment of meningitis. Experience with penicillin in the treatment of bacterial meningitis reported elsewhere [4] indicated that parenteral therapy suffices and that intrathecal injection is unnecessary.

CASE 6.—J. M., aged 15, a patient of Dr. W. Daiber, complained of headache about April 7 which became much worse, with fever and signs of meningitis. The spinal fluid on two occasions was under pressure of 600 mm. of water, contained 200 leukocytes, mostly lymphocytes; 44 and 30 mg. of sugar, and 640 and 600 mg. of chloride. A pellicle formed, and in one specimen acid-fast rods were reported. Cultures of the spinal fluid remained sterile. In later samples the amount of sugar present was too small to measure.

On the twenty-fifth day of disease streptomycin was given by continuous intravenous drip, 4 Gm. daily, for seven days. No significant changes occurred in the spinal fluid, somnolence deepened into coma and death occurred on the thirty-second day. The diagnosis was confirmed at necropsy. During therapy the amount of streptomycin in the spinal fluid measured on five occasions varied from 16 to 20 micrograms per cubic centimeter.

TABLE 1.—*Amounts of Streptomycin in the Spinal Fluid after Various Parenteral Doses Compared with Amounts in the Blood*

Case	Daily Dose, Gm.	Micrograms per Cc. of Spinal Fluid	Micrograms per Cc. of Blood
O. G.*	1, intramuscularly	5 (estimated)	5 (estimated)
9. J. S.	1, intravenously	4 (estimated)	
1. H. B.	4, intravenously	20	12
2. L. B.	4, intravenously	5 (estimated)	20
3. C. M.	5, intravenously	11	30
	5, intravenously	6 (estimated)	14
6. J. M. (tuberculous meningitis)	4, intravenously	16 20 18 18 18	19

* Reimann, Elias and Price,[1] case 1.

EFFECT OF STREPTOMYCIN ON THE FLORA OF THE URINE

The fact that from 40 to 60 per cent of streptomycin injected parenterally is excreted in the urine where it appears in bactericidal amounts, suggests its therapeutic trial in cases of infection of the urinary tract caused by gram-negative bacilli. Plans were made accordingly to study the effect of streptomycin on the bacteria in infected urine. Intramuscular or intravenous injection of between 1 Gm. and 5 Gm. causes 150 to 2,800 micrograms of streptomycin per cubic centimeter to appear in urine, depending on the volume of urine (table 2). In this respect, intramuscular therapy and a restricted intake of fluid, to obtain a relatively large amount of drug in a small amount of urine,

4. Price, A. H., and Hodges, J. H.: Treatment of Meningitis with Penicillin Injected Intravenously and Intramuscularly, New York State J. Med. **44**:2012-2014 (Sept. 15) 1944.

is usually preferable to intravenous therapy, which requires the daily injection of 3,000 cc. of fluid. However, in 1 patient the intravenous injection of 5 Gm. resulted in 2,800 micrograms of streptomycin per cubic centimeter of urine, the highest amount measured. Considerable daily variation occurs even with constant dosage, as indicated by sample readings shown in table 2.

TABLE 2.—*Variations in Amount of Streptomycin per Cubic Centimeter of Urine in Relation to Dosage, Route of Injection and Amount of Urine Excreted*

Case	Daily Dose, Gm.	Micrograms per Cc. of Urine	Daily Urinary Output, Cc.
3. C. M.	1, intramuscularly	150	2,650
		280	2,800
	3, intravenously	540	4,300
		2,500	2,780
	5, intravenously	2,800	3,440
10. J. W.	1, intramuscularly	300	1,200
	2, intramuscularly	1,700	1,300
9. J. S.	1, intramuscularly	115	1,100
	1, intravenously	150	3,600
7. B. B.	2, intravenously	310	3,800
		640	3,000
4. G. B.	4, intravenously	700	3,100
		1,630	810
		700	3,700

Amounts accumulate rapidly in the urine within a few hours after beginning treatment and disappear about forty-eight hours after stopping it.

BACILLUS PYOCYANEUS INFECTION

CASE 7.—Mrs. B. B., aged 32, had hydronephrosis and pyuria, with gram-negative rods and gram-negative cocci in the urine. On culture only colonies of Bacillus pyocyaneus grew. Shortly after operation for dilation of the urethra and urethral orifices, the temperature rose to 106 F. and the leukocytes to 12,000 per cubic millimeter of blood. Streptomycin was given by the intravenous drip method in doses of 2 Gm. daily for four days. During this time the temperature rapidly declined to normal. Before treatment the number of B. pyocyaneus organisms estimated in the urine was 2,000,000; forty-eight hours after the beginning of treatment, the number was reduced to 2,000 and on the fourth day to 1,500. The amount of streptomycin in the urine varied from 300 to 640 micrograms per cubic centimeter, representing the usual 40 to 60 per cent recovery of parenterally injected streptomycin. Growth of the B. pyocyaneus involved was inhibited on agar containing 100 micrograms per cubic centimeter but not 75 micrograms.

CASE 8.—C. G., a debilitated man aged 66, studied in collaboration with Dr. John Eimann at Abington Memorial Hospital, had a severe infection of the urinary tract with B. pyocyaneus and Bacillus proteus morganii. He was given 4 Gm. daily intravenously for five days, but edema developed and the intramuscular route was chosen. His general condition became worse, and treatment was stopped.

Culture of the urine during treatment showed a disappearance of Proteus morganii, which was inhibited by 8 micrograms per cubic centimeter of agar, but B. pyocyaneus which was on test was resistant to more than 200 micrograms per cubic centimeter. Evidently different strains of B. pyocyaneus also vary in their resistance to streptomycin.

EBERTHELLA INFECTIONS

CASE 9.—J. S., a man aged 48, suffered from chronic infection of the urinary tract after a pelvic injury four years previously. He complained of frequent burning, and his urine was often cloudy. A stained smear showed a mixture of bacteria, mostly gram-negative rods, and variable other bacilli, which on culture were E. coli (predominant), Klebsiella pneumoniae and a hemolytic gram-positive rod. The numerical distribution of bacteria before and after treatment with streptomycin is shown in table 3. The colon bacillus isolated was inhibited by 3 micrograms but not by 2 in the test tube.

The patient's urinary output was diminished by limiting the fluid intake to 1,000 cc. per day in order to concentrate the excreted streptomycin. The drug was given intramuscularly in doses of about 0.125 Gm. every three hours, or 1 Gm. daily. Pain at the site of injection and slight headache occurred shortly after the first dose. Each of the next five injections was followed by severe headache lasting about thirty minutes. After two days of intramuscular therapy because of headache, the same dosage was given by the intravenous route for four days. No further headache occurred, probably because of more gradual administration of the drug. The effect on the bacterial count is shown in table 3. Within twenty-four hours urine was free from all bacteria, including the gram-positive bacilli, except anaerobic ones.

Tests were made to determine the lasting effect of therapy on the urinary flora. Two days after treatment was stopped, a trace of streptomycin was still present in the urine to account for the persistent absence of certain unidentified aerobic bacteria, yet when the urine was diluted to reduce the amount of drug present, the hemolytic bacilli grew, indicating that streptomycin had a suppressive but not a bactericidal effect. No colon bacilli grew at any dilution. Seven days after treatment was stopped neither colon bacilli nor the hemolytic bacilli grew on the culture plates.

The spinal fluid contained 4 micrograms of streptomycin per cubic centimeter with doses of 1 Gm. daily intravenously.

CASE 10.—J. W., a man aged 65, had urinary retention complicated by prostatic hypertrophy and obstruction. A retention catheter was inserted for drainage. The urine was purulent and contained a variety of bacteria as indicated in table 4. The colon bacillus present was inhibited by 6 micrograms of streptomycin but not by 5 micrograms.

The fluid intake was restricted to 1,200 cc. a day, and streptomycin was given intramuscularly in doses of 2 Gm. for two days. Injections were followed immediately by pain locally and moderate headache lasting thirty minutes accompanied with a temporary fall of blood pressure from 128 systolic and 78 diastolic to 110 systolic and 40 diastolic and once to 90 systolic and 40 diastolic. The dosage was reduced to 1 Gm. for four days with only occasional mild headache after injection. The effect of therapy on the bacteria in the urine is illustrated in table 4. As in case 9, within twenty-four hours the urine was cleared of all bacteria except anaerobic ones.

Higher dilutions of urine made from the sample of the third day of treatment gave growth of a few colonies

of various unidentified aerobic bacteria, indicating that the streptomycin present in undiluted urine inhibited their growth but when it was further diluted growth occurred. E. coli failed to grow in any dilution.

Samples tested two days after treatment was stopped still contained traces of streptomycin, and the total aerobic bacterial count increased to 32,000,000, but colon bacilli were absent. Seven days later colon bacilli and hemolytic cocci were both absent.

In 1 patient, 4 Gm. of streptomycin given intravenously failed to eliminate E. pyogenes from the urine, although 1,500 units were present in it.

EFFECT OF STREPTOMYCIN ON FLORA OF THE FECES

Streptomycin given orally is almost all excreted in the feces. In studies in a case of typhoid (Reimann, Elias and Price [1] case 5) oral administration of 4 Gm. daily caused from 10,000 to 20,000 micrograms of streptomycin to appear per gram of the feces and eliminated both E. typhosa and E. coli from the feces during treatment, giving an almost odorless stool. Both varieties of bacilli reappeared several days after oral therapy was stopped, when only 40 micrograms of streptomycin per gram of feces remained. It was suggested that for intestinal infections susceptible to the effects of streptomycin, the drug should be given parenterally to control the systemic infection and orally to sterilize the feces and prevent reinfection.

The results also suggested that streptomycin may be of value to reduce the number of susceptible bacteria in the intestine prior to surgical operation. With this in mind the following patients were treated and studied.

Streptomycin was given orally every three hours to each patient, the amount depending on the total daily dose decided on.

CASE 11.—Mrs. G. S. was given 2 Gm. of streptomycin by mouth daily in doses of 0.25 Gm. every three hours for three days before operation for carcinoma of the colon with subacute obstruction. Bacterial counts made before treatment showed 1,300,000 organisms on nutrient agar, 16,000,000 in anaerobic culture medium and 10,000 colon bacilli on MacConkey's medium. Feces passed during the first twenty-four hours of therapy contained 650 micrograms of streptomycin per gram; the number of bacteria on nutrient agar and on culture medium for anaerobes was unchanged, but colon bacilli were absent. Stools passed during the second twenty-four hours contained 850 micrograms per gram with the bacterial growth about the same; no colon bacilli grew.

CASE 12.—Mrs. R. H. was given the same daily dosage of streptomycin as in case 11 for two days before operation for carcinoma of the colon with subacute obstruction. Before therapy too many bacteria to count grew on all three mediums. After twenty-four hours of treatment the stools contained 3,200 micrograms per gram, with a reduction to 1,000,000 colonies on nutrient agar and slight reduction, to 100,000,000 on anaerobic mediums, but no colon bacilli grew. The next day, with 600 micrograms per gram, the count was 400,000 on nutrient medium; there was no reduction in anaerobic culture and again no colon bacilli.

In these 2 patients, 600 micrograms of streptomycin per gram of feces was enough to eliminate colon bacilli. At operation the surgeons found the bowel not distended and well prepared for operation.

CASE 13.—Mrs. K. S., aged 65, a diabetic patient with carcinoma of the colon, was given 1 Gm. orally for seven days. The amount of streptomycin in the feces varied from 1,000 to 5,800 micrograms per gram but was without effect on the numbers of any bacteria, including E. coli, as compared with a sample plated

TABLE 3 (case 9, J. S.).—*Effect of Parenteral Therapy on Urinary Flora*

Day	Dose, Gm.	Urine Volume, Cc.	Micrograms per Cc.	Total Aerobic Count	Coliform Bacilli	Hemolytic Bacilli	Anaerobic Bacteria
Before treatment		.....	...	5,000,000	600,000	500,000	700,000
1	1 intramuscularly	1,100	115	0	0	0	Present
3	1 intravenously	980	770	0	0	0	Present
4	1 intravenously	2,170	120	0	0	0	Present
6	1 intravenously	3,650	150	3,200	0	..	Present
Two days after treatment		.....	5	2,000,000	0	0	Very few present
Nine days after treatment		.....	None	2,000,000	0	0	

TABLE 4 (case 10, J. W.).—*Effect of Parenteral Therapy on Flora of the Urine*

Day	Dose, Gm.	Urine Volume, Cc.	Micrograms per Cc.	Total Aerobic Count	E. Coli	Hemolytic Cocci	Anaerobic Bacteria
Before treatment		.....	...	7,000,000	5,000,000	2,000,000	1,000,000
1	2 intramuscularly	1,450	150	0	0	0	Present
2	2 intramuscularly	1,320	1,700	0	0	0	0
3	1 intramuscularly	2,600	300	0	0	0	Present
6	1 intramuscularly	1,200	300	18,000,000	0	..	Present
8	2 days after treatment	.....	13	32,000,000	0	8,000	Present
15	9 days after treatment	.....	None	8,000	0	0	Present

before treatment. Dosage was increased to 2, 3, 4, 5 and finally 6 Gm. at intervals of several days. At 5 Gm. the amount of streptomycin rose to 16,000 micrograms per gram but again without significant influence on the numbers of bacteria, as shown in table 5.

Only after the dose of 6 Gm. were the counts significantly reduced. The failure to eliminate these colon bacilli was perhaps accounted for by their resistance to streptomycin; they grew on agar containing 200 micrograms of streptomycin per cubic centimeter, while growth of colon bacilli in cases 11 and 12 was inhibited by 6 micrograms.

CASE 14.—Mrs. B. E., aged 65, with carcinoma of the colon, had a count of 4,000,000 aerobic bacteria and 3,000,000 E. coli. She received 5 Gm. of streptomycin daily for two days. After twenty-four hours of treatment no aerobic bacteria and no colon bacilli grew. Anaerobic bacteria were unaffected. On the first and second days 400 micrograms and 760 micrograms of streptomycin respectively were present per gram of feces.

CASE 15.—E. B., with carcinoma of the colon, had 10,000,000 anaerobic bacteria and 6,000,000 E. coli per gram of feces. He was given 5 Gm. of streptomycin orally for five days. After forty-eight hours of treatment the number of aerobic organisms was reduced to 50,000 and colon bacilli to 0, while 3,300 micrograms of streptomycin was present per gram of feces.

CASE 16.—D. R., with carcinoma of the colon, before treatment had 1,000,000,000 each of aerobic bacteria and E. coli per gram in his feces. After one day of 5 Gm. of streptomycin, only 10,000 aerobic bacteria and no colon bacilli remained in the feces, which contained 6,500 micrograms of streptomycin per gram.

The surgical aspects of the treatment and its effect in these patients will be the subject of a separate report.

Streptomycin was given orally in doses of 5 Gm. daily for seven daily to a patient with severe ulcerative colitis. During therapy the number of stools lessened but the fever persisted unchanged.

TABLE 5 (case 13, K. S.).—*Effect of Oral Therapy on Resistant Bacteria in the Feces*

Days	Oral Dosage, Gm.	Weight Feces, Gm.	Micrograms per Gm. Feces	Nutrient Medium	MacConkey's Medium	Anaerobic Bacteria
Pretreatment.....	.....	...	.....	41,000,000	27,000,000	Uncountable
3.....	1	182	2,700	56,000,000	70,000	
7.....	1	41	5,800	270,000,000	115,000,000	80,000,000
9.....	2	258	9,100	170,000,000	125,000,000	16,000,000
14.....	4	289	3,700	315,000,000	78,000,000	200,000,000
15.....	5	57	16,400	400,000,000	100,000,000	Many
16.....	5	238	5,200	300,000,000	120,000,000	Many
18.....	6	36	1,900	1,000,000	250,000	Present

TABLE 6.—*Resistance to Streptomycin of Bacteria Isolated During this Study*

Case	Strain	Micrograms of Streptomycin Not Inhibitive	Micrograms of Streptomycin Which Inhibit
	Stock, E. typhosa....	..	1
Epidemic*	E. typhosa....	4	6
1. H. B.	E. typhosa....	7	9
2. L. B.	E. typhosa....	16	18
3. C. M.	Br. suis.....	0.1	0.5
4. G. B.	Br. suis.....	0.1	0.5
5. A. A.	Br. suis.....	0.1	0.5
7. B. B.	B. pyocyaneus	75	100
8. C. G.	B. pyocyaneus	>200	
	P. morganii...	7	8
9. J. S.	E. coli.....	2	3
10. J. W.	E. coli.....	5	6
13. K. S.	E. coli.....	>200	

* Reimann, Elias and Price.[1]

TOXICITY

In these studies the only evidence of toxicity of streptomycin was transient but severe headache in 2 patients treated intramuscularly (cases 9 and 10), headache and a fall in blood pressure in 1 and erythema in 1 treated intravenously (case 3).

COMMENT

There is reason to believe that streptomycin holds promise as a therapeutic agent for systemic infections caused by certain gram-negative, streptomycin-sensitive bacilli which are not influenced by penicillin or sulfonamide compounds, since the amounts attained in the body by parenteral or oral administration often exceed the amounts needed to kill these bacilli on agar. Yet for infections caused by relatively less sensitive bacilli, like E. typhosa, the clinical results thus far reported are not as satisfactory as desired. No benefit occurred in some instances in which the amount of streptomycin in the blood greatly exceeded the theoretic bactericidal level early in the disease (case 1), and in 1 instance (case 2) an apparent good result was obtained in infection with a strain of E. typhosa which resisted 16 micrograms of streptomycin in vitro. The matter is further complicated by evidence of considerable variation in resistance to streptomycin (1 to 18 micrograms per cubic centimeter) of different strains of E. typhosa; by irregularities in the amounts of streptomycin attained in the body with similar dosage in the same patient and in different patients; by the possibility that intracellular bacteria may not be affected by the drug; by the possibility of the development of drug resistance by the bacteria during treatment; by the possibility that the hydrogen ion concentration of autolysing tissues and pus may be low enough to inactivate basic

streptomycin,[5] and by the probable variation in the lots of streptomycin available at present. In certain instances as much streptomycin was measured in the blood after the injection of 1 Gm. intramuscularly as with 5 Gm. given intravenously, and increases of dosage were not always followed by proportionate increments in the blood. Amounts in the blood did not exceed 27 micrograms per cubic centimeter in any of the patients studied. It is evident that many more patients with typhoid treated with streptomycin must be studied before its value can finally be assessed.

Even less can be said at present for the treatment of brucellosis from studies in only 3 cases. Despite the fact that Br. suis obtained from patient 3 was far more sensitive to streptomycin in vitro than E. typhosa, no beneficial effect was apparent until sixty times the theoretic amount needed was attained in the blood. Even then, the forty-eighth day of disease, a spontaneous remission may have clouded the issue. In case 4 recovery may also have been spontaneous, but it appears very likely that the disease was terminated by streptomycin, although amounts of only 19 micrograms per gram of blood were attained. In case 5 no beneficial effect occurred, although the patient for her size received more streptomycin per kilogram than any other one. Similar uncertain results were reported by Herrell and Nichols.[1g]

No benefit occurred in the patient with tuberculous meningitis in whom as much as 20 micrograms per cubic centimeter appeared in the spinal fluid. Nevertheless, amounts of streptomycin theoretically inhibitive for certain bacilli do appear in spinal fluid during parenteral therapy.

Streptomycin was an effective agent for reducing the numbers of or eliminating certain gram-negative bacilli from the urine in several patients when it was injected parenterally, but, again, the problems of the irregular amounts of drug which appear in urine and of variability of resistance of strains even of the same genus of bacteria arise. Of two strains of B. pyocyaneus, the one which was not eliminated by therapy was more than twice as resistant to the drug as the other. In 2 other patients E. coli, and in a third B. proteus morganii, were promptly eliminated with ease. The resistance to streptomycin on agar of various bacteria from patients in this study is presented in table 6.

5. Florey, H. W.: The Use of Micro-Organisms for Therapeutic Purposes, Brit. M. J. **2**:635-642 (Nov. 10) 1945.

Streptomycin given orally promptly suppresses or eliminates drug-sensitive strains of E. coli and E. typhosa and certain other bacteria from the feces so long as an adequate amount is maintained therein. Cessation of treatment is followed by rapid return of the flora, as may be expected, but even a temporary suppression is of value at times, particularly during intestinal operations. Here again in the few patients studied there was evidence of great variability in the resistance of different strains of E. coli to streptomycin; in most cases they were suppressed or temporarily eliminated, but in 1 diabetic patient a strain resistant to 200 micrograms per cubic centimeter of agar (the highest concentration tested) persisted in the stool despite the presence of 16,000 micrograms per gram of feces. Besides variation in bacterial resistance and in hydrogen ion concentration the matter of uniform admixture of streptomycin with feces must be considered. For better distribution, small doses fed often are probably better than large doses at longer intervals. No information is available about the amount of streptomycin which adheres to the wall of the bowel and of its effects on bacteria in the crypts or on the mucosal surface.

The relative value of streptomycin and other urinary and intestinal tract antiseptics, such as mandelic acid, sulfonamide compounds and penicillin, will have to be determined by further work. It can be said, however, that streptomycin is probably much less apt to cause toxic effects than the sulfonamide compounds. It may approach even closer to being the ideal intestinal antiseptic than ones now used since it is poorly absorbed from the digestive tract, it is nontoxic and eliminates certain pathogenic gram-negative bacteria from the feces.

SUMMARY

Streptomycin seemed to be of value in the treatment of 6 of 10 patients with typhoid reported on in this and in a previous study [1a]; no improvement from therapy occurred in 2, and 2 died. Different strains of typhoid bacilli vary in their resistance to streptomycin. There is evidence also that certain factors in the body inhibit the action of the drug or protect the bacteria from its effects.

The results of treatment of 3 patients with brucellosis are inconclusive. Recovery seemed related to therapy in 1 and doubtfully related in a second, and no benefit occurred in the third. No effect was noted against tuberculous meningitis, but streptomycin appears in the spinal fluid in theoretically bacteriostatic amounts for

certain bacteria when injected parenterally, whether the meninges are normal or diseased.

Streptomycin in adequate dosage given parenterally promptly suppressed or eliminated B. pyocyaneus, B. proteus morganii and E. coli from the urine of several patients but only when these bacteria were of strains susceptible to the amounts of streptomycin present in the urine.

Streptomycin given orally promptly suppressed or eliminated E. coli and certain other bacteria in the feces during therapy but failed to do so when the bacteria were resistant to the amounts of drug present.

With similar dosages, irregularities in the amounts of streptomycin in the blood, urine, spinal fluid and feces occur in the same patient and in different patients. Within limits, the sensitivity of certain bacteria to streptomycin in culture mediums was not a reliable guide to the amount needed to suppress them in the body, but with highly resistant forms, and occasionally with susceptible forms, therapy was ineffective. It is probable that certain factors in the body interfere with the bacteriostatic action of streptomycin.

HEMOLYTIC STREPTOCOCCUS SORE THROAT

DETAILED STUDY OF THE SIMULTANEOUS INFECTION OF A LARGE NUMBER OF MEN BY A SINGLE TYPE

LOWELL A. RANTZ, M.D.; WESLEY W. SPINK, M.D., AND PAUL J. BOISVERT, M.D.
SAN FRANCISCO MINNEAPOLIS NEW HAVEN, CONN.

Clinicians interested in infectious disease are aware that the exposure of a group of human beings or animals to an infectious agent will be followed by the development of clinical syndromes of varying degrees of severity. It is rarely possible to describe these phenomena in detail in human beings, since artificial inoculation is infrequently performed and since natural infection is usually unsuitable because the size of the infecting dose and the strain of microorganism may be expected to vary from case to case. These difficulties are particularly evident in the study of group A hemolytic streptococcus infections, since many serologic types are known to occur among these organisms.

One of us has previously described [1] certain clinical and immunologic aspects of the disease resulting from the simultaneous infection of a large group of men by a single type of hemolytic streptococcus. Clinical observations in that epidemic were incomplete. Recently another opportunity for the study of a similar outbreak presented itself. This was a food-borne epidemic of type 1 hemolytic streptococcus sore throat which occurred among the patients of a large station hospital. More than 250 cases of clinical infection were discovered. A man working in the kitchen who was a carrier of type 1 streptococci in the nose and throat was believed to be the primary source of the infectious agent. On this occasion a smaller group of men was selected and studied in detail.

The laboratories of the Department of Medicine, Stanford University School of Medicine, were made available to the commission for certain studies.

This investigation was carried out during a field study by the Commission on Hemolytic Streptococcal Infections, Board for the Investigation and Control of Influenza and Other Epidemic Diseases in the Army, Preventive Medicine Service, Office of the Surgeon General, United States Army.

1. (*a*) Bloomfield, A. L., and Rantz, L. A.: An Outbreak of Streptococcic Sore Throat in an Army Camp, J. A. M. A. **121**:315 (Jan. 30) 1943. (*b*) Rantz, L. A.: Group A Hemolytic Streptococcus Antibodies: III. A Study of the Simultaneous Infection of a Large Number of Men by a Single Type, Arch. Int. Med. **73**:238 (March) 1944.

MATERIALS AND METHODS

Fifty-eight hospital patients, all suffering from functional gastrointestinal disease, were selected for study. They were particularly suitable for this purpose, since a complete laboratory work-up, including leukocyte count and erythrocyte sedimentation rate, had been previously performed in each case. An analysis of their diets indicated that all had been equally exposed to infection. The first clinical evidence of the outbreak was noted on April 16, 1944. Each of the study group was carefully examined and materials from the throat obtained for culture on April 17 and 18. Those who were found to harbor hemolytic streptococci in the nasopharynx were accepted for more detailed study, including close clinical observation and serial cultures of materials from the throat, Dick tests, leukocyte counts, measurements of erythrocyte sedimentation rate (Westergren) and antifibrinolysin [2] and antistreptolysin titrations [3] over a period of twenty-four to thirty days. The isolated hemolytic streptococci were grouped and typed by the precipitin technics of Lancefield.[4]

RESULTS

Group A hemolytic streptococci were discovered on the initial examination in the throats of 40 of the 58 men in the study group. In 5, the organisms were not strains of the epidemic type, and these patients were not further considered. Thirty-five persons had presumably become carriers of type 1 organisms as a result of exposure to infected food.

It was not possible to determine all the factors that prevented the epidemic type from gaining lodgment in the throats of 40 per cent of these men, but the most important factor was the absence of tonsils. These organs had been removed from 12, or 52 per cent, of the uninfected but from only 2, or 5.7 per cent, of the infected men.

2. Boisvert, P. J.: The Streptococcal Antifibrinolysin Test in Clinical Use, J. Clin. Investigation **19**:65, 1940.

3. Rantz, L. A., and Randall, E.: A Modification of the Antistreptolysin Determination, Proc. Soc. Exper. Biol. & Med. **59**:22 (1945).

4. Lancefield, R. C.: A Serological Differentiation of Human and Other Groups of Hemolytic Streptococci, J. Exper. Med. **57**:571, 1933. Swift, H. F.; Wilson, A. T., and Lancefield, R. C.: Typing Group A Hemolytic Streptococci by M Precipitin Reactions in Capillary Pipettes, ibid. **78**:127, 1943.

TABLE 1.—*Summary of Clinical, Clinicopathologic and Immunologic Observations on Thirty-Five Men Who Became Carriers of Type 1 Hemolytic Streptococci*

Maximum Temperature, Nearest Fahr. Degree	Number of Cases	Subjective Sore Throat		Tonsillar Exudate		Tender Cervical Adenitis		Average Duration Fever, Days	Average Duration Objective Signs, Days	Total Leukocyte Count				Erythrocyte Sedimentation Rate (Westergren)				Antistreptolysin Titer					
										Range Highest Counts, × 1,000	Average Maximum Count, × 1,000	Significant Increase		Range Most Rapid Rates, Mm./Hr.	Average Maximum, Mm./Hr.	Significant Increase		Positive Dick Test, Number	Average Initial, Units/Cc.	Average Maximum, Units/Cc.	Average Fold, Increase	Significant Increase	
		No.	%	No.	%	No.	%					No.	%			No.	%					No.	%
103+	8	8	100.0	8	100.0	7	87.5	2.6	4.4	14-20	17.3	8	100.0	7-43	39	7	87.5	0	97	250	2.5	7	100.0
102	6	6	100.0	4	66.6	5	83.4	2.5	3.3	7-20	14.5	5	83.4	5-26	12	4	66.6	1	160	284	1.8	5	83.4
101	5	5	100.0	5	100.0	4	80.0	2.4	3.0	9-21	16.8	3	60.0	4-35	15	2	40.0	2	118	145	1.2	1*	33.3
100	5	3	60.0	4	80.0	3	60.0	1.6	4.0	11-15	13.0	5	100.0	3-18	8	0	0.0	1	120	180	1.5	4	80.0
99	6	4	66.6	4	66.6	3	50.0	1.2	2.3	8-15	10.8	2	33.3	1-68	18	3	50.0	3	68	110	1.9	4	80.0
98.6	5	2	40.0	1	20.0	1	20.0	0.0	1.2	9-18	12.4	1	20.0	1-22	7	1	20.0	0	105	192	1.8	2	40.0

* Two were followed for less than one week.

Streptococci of type 1 were present in the throat in large numbers for at least one week in all but 2 patients. A persistent carrier state may, therefore, be said to have been established.

Clinical Observations.—Disease of widely varying degrees of severity occurred among the men infected by the epidemic streptococcus. The pertinent data are summarized in table 1. The cases have been divided into groups on the basis of the nearest whole Fahrenheit degree of the maximum recorded temperature. Subjective sore throat was present in every patient whose temperature was greater than 100 F., in 60 per cent of those with lesser degrees of fever and in 2 of the 5 whose temperatures remained normal throughout.

Tonsillar and pharyngeal exudate and tender anterior cervical adenitis have been the two most valuable signs for the clinical recognition of hemolytic streptococcus sore throat. The frequency of their occurrence in this group was roughly correlated with the height of fever, both being present more frequently and in a more definite form in patients whose temperature reached 101 F. One or the other of these signs was definitely present in all but 2 of the patients in whom a rise in temperature occurred. The latter men were definitely infected, since an antistreptolysin response was demonstrated during convalescence. Either exudate or adenitis was absent in several of the most severely ill patients, in all of whom the tonsils were intact. In others these signs were discovered only by the most careful examination.

The disease in the definitely febrile patients ran a short course, the temperature being elevated for an average of from two and five-tenths days in the most severely ill to one and two-tenths days in the least sick. The objective signs in the throat and the adenitis persisted for one or two days after the fever had disappeared.

The total leukocyte count on the second day of the disease was increased in nearly all febrile patients and in a few of the afebrile ones. The number was correlated with the maximum temperature, but a few men with a definite febrile illness failed to respond to the infection with an increase of these cells in the blood. The rate of erythrocyte sedimentation on the second day of illness was elevated for many patients and was greatest for those patients with temperatures above 103 F. A significant increase of 10 mm. per hour or more was observed in about half of all patients and in 4 of the 11 in whom little or no fever was detected.

Antibody Response.—An increase in the amount of antifibrinolysin during convalescence was observed in only 1 case. This is in accord

with data obtained in other cases of type 1 infections and may be correlated with the weak fibrinolysin produced in vitro by these organisms.

Seven of the 35 patients who became carriers of type 1 organisms and in whom definite evidence of infection was obtained had positive Dick reactions. In none did a rash develop, and cutaneous sensitivity was not lost during convalescence, indicating that antitoxin had not been formed in response to the infectious process.

The serum antistreptolysin titer was measured on the second day of the illness and during convalescence. There was no correlation between the initial level of this antibody and the nature and severity of the clinical disease. The average final titer was somewhat higher in the more severely ill patients, but such an observation is without meaning unless it is correlated with the initial titer, as has been discussed elsewhere.[11b] The ratio, $\frac{\text{maximum titer}}{\text{initial titer}}$, is a convenient value which expresses this relationship. A ratio of 1.5 indicates that a significant antibody response has ensued.

Such an increase in circulating antistreptolysin occurred in 66 per cent of the 35 men who became carriers of type 1 organisms and was slightly greater in magnitude and frequency in severely ill patients. It was also observed after infection associated with no fever or with only a slight febrile reaction. Exudate and adenitis were completely absent in 3, and minimal in 4, in which an antibody response was discovered. Four patients were febrile, and leukocytosis was demonstrated in each.

Four other febrile patients failed to exhibit an antistreptolysin response. Tonsillar exudate and cervical adenitis were minimal but definite in 3 and extensive in the fourth. A significant elevation in the leukocyte count and erythrocyte sedimentation rate was observed in 3.

Low Grade Infection.—The clinical course of infection by type 1 hemolytic streptococci, as observed during a food-borne epidemic of sore throat, has been described and the great variation in the clinical course of the disease emphasized. Serious nonsuppurative complications frequently are sequelae of acute streptococcic disease of the respiratory tract, and, because it is believed that these processes may follow even the mildest infection, it is worth while to describe in greater detail the nature of the disease observed in 11 of the study group in whom the temperature was never greater than 99 F. The essential data are presented in table 2.

Malaise was minimal in all of these men, but definite subjective sore throat was present in 6. Exudate and/or adenitis of the cervical lymph nodes was discovered on physical examination of 7 men, including 2 whose throats were not subjectively sore. Symptoms were lacking, and the physical examinations simultaneously revealed nothing significant in only 3 cases.

TABLE 2.—*Summary of Clinical, Clinicopathologic and Immunologic Observations on Eleven Men Who Became Carriers of Type 1 Hemolytic Streptococci and in Whom Minimal Systemic Reaction Occurred*

Patient	Maximum Temperature	Subjective Sore Throat	Tonsillar Exudate	Tender Cervical Adenitis	Total Leukocyte Count × 1,000		Erythrocyte Sedimentation Rate, Mm./Hr.		Antistreptolysin Titer		
					Before Infection	Maximum	Before Infection	Maximum	Onset Infection, Units/Cc.	Maximum,† Units/Cc.	Fold Increase
1	99	Yes	++	++	5	8	3	19	50	125	2.5
2	99	Yes	+	0	5	10	2	11	50	50	0.0
3	99	No	+	0	10	10	15	68	50	100	2.0
4	99	Yes	0	+	9	15	1	7	50	125	2.5
5	99	Yes	0	++	7	10	2	1	50	166	3.3
6	99	No	++	0	9	12	4	4	100	100	0.0
7	98.6	Yes	+++	+++	7	12	2	7	10	50	5.0
8	98.6	Yes	0	0	10	11	8	22	166	625	3.7
9	98.6	No	0	0	8	9	1	1	166	166	0.0
10	98.6	No	0	0	12	12	2	2	125	125	0.0
11	98.6	No	0	0	10	18	2	2	50	50	0.0

+ indicates minimal exudate or adenitis; ++, definite easily recognizable exudate or moderately severe adenitis; +++, very large amounts of exudate.

The tissue reaction to infection, as measured by the increase in total leukocyte count and erythrocyte sedimentation rate, was minimal in this group of low grade infections. The latter value was elevated for 3 patients; for 1 presenting practically no other signs of infection, to an extreme degree.

A significant increase in the serum antistreptolysin titer occurred during convalescence in 6 cases, and in 2 others there were definite clinical signs of infection. Clinical and/or immunologic evidence of tissue reaction to the epidemic streptococcus was lacking in only 3 of the whole group of 35 who became carriers, and a sharp increase in the total leukocyte count occurred in 1 of the latter.

Complications.—Suppurative and nonsuppurative complications were notably infrequent in

men infected during the outbreak. It is particularly important that rheumatic fever was never a sequel to the disease, although more than 250 infections were recognized clinically throughout the hospital.

Reinfection.—Several hundred patients were resident in the hospital at the time of the epidemic, of whom 60 (in three wards) had group A hemolytic streptococcus sore throat due to a variety of serologic types. These patients were in various stages of convalescence and had previously been under observation. Forty-four were adequately studied after the epidemic, the others having been discharged from the hospital. Type 1 streptococci were isolated from the throats of 11 in whom this type had not previously been present. Sore throat and cervical adenitis, but not exudate, were noted in 3 of this small group. In 4 other patients an elevation in the erythrocyte sedimentation rate followed exposure to the epidemic streptococci.

Type 1 streptococci were never recovered from the nasopharynxes of 33 patients in the postepidemic period. This does not establish the fact that they had not gained lodgment there, since most of these men were already carriers of a different type and the selective isolation of a new strain under these circumstances is difficult. The patients were, however, exposed to reinfection. A slight increase in temperature occurred in 3, in association with sore throat in 2. Cervical adenitis developed in none. In spite of these minimal clinical signs, an increase in total leukocyte count and/or erythrocyte sedimentation rate was demonstrated in 15 during the postepidemic period.

Minimal signs of infection were noted in only 6 of this group of 44 patients, but laboratory evidence of a tissue reaction to the different type of streptococcus in the absence of clinically demonstrable disease was discovered in 19. These observations assume greater significance when it is realized that definite, clinically recognizable hemolytic streptococcic sore throat developed in 50 to 60 per cent of the remaining hospital patients who had not previously undergone an infection by group A streptococci.

COMMENT

Information has been accumulating for more than fifteen years [5] which has indicated that group A hemolytic streptococcus infection of the respiratory tract is intimately associated with the development of rheumatic fever. Recent definitive studies [6] show that this serious disorder occurs only as a sequel to infection by these organisms and, furthermore, that a potentially serious nonarthritic continuing disease process is initiated by such infection in many persons. It has, therefore, become important to describe the natural history of hemolytic streptococcic disease in detail. Comparative studies of natural infection are difficult, because the size of the infecting dose and strain of the etiologic agent may vary from case to case. This paper summarizes the results of clinical and laboratory observations of a food-borne epidemic of type 1 hemolytic streptococcus sore throat in a military hospital. Under these circumstances it is reasonable to assume that the same organism, ingested by each man in approximately comparable numbers, was responsible for the outbreak.

Fifty-eight men, of whom 35 became carriers of the epidemic type, were studied clinically and bacteriologically. The others may not have consumed the contaminated food, but another factor appears to have been at work. Tonsillectomy had been performed approximately 9 times as frequently among those men who did not become carriers as among those who did. This observation is to be contrasted with those made in the study of another large group of cases, in which the infection was caused by a variety of types of the organism and in which the absence of tonsils was not found to interfere with the development of streptococcic sore throat.[7] Bloomfield [8] has previously noted the protection offered by tonsillectomy against infection by streptococci, but the observations recorded here indicate that the nature and type of the etiologic agent must be carefully defined before such studies can be compared and evaluated.

The clinical disease which occurred among the 35 men who became carriers of the epidemic type varied greatly, and the cases could be divided into groups on the basis of maximum temperature. When the temperature reached 101 F. or more, the disease was usually "typical," in that sore throat, tonsillar exudate, adenitis involving the anterior cervical lymph nodes, with tender-

5. Paul, J. R., and others: The Epidemiology of Rheumatic Fever and Some of Its Public Health Aspects, New York, Metropolitan Life Insurance Company for the American Heart Association, 1943.

6. Rantz, L. A.; Boisvert, P. J., and Spink, W. W.: Etiology and Pathogenesis of Rheumatic Fever, Arch. Int. Med. **76**:131 (Sept.) 1945.

7. Rantz, L. A.; Spink, W. W., and Boisvert, P. J.: Hemolytic Streptococcal Sore Throat: The Acute Disease, to be published.

8. Bloomfield, A. L., and Felty, A. R.: Bacteriologic Observations on Acute Tonsillitis with Reference to Epidemiology and Susceptibility, Arch. Int. Med. **32**: 483 (Oct.) 1923.

ness, leukocytosis and increased erythrocyte sedimentation rate were observed. Either tonsillar exudate or cervical lymphadenitis or both, was present in all but 2 of the febrile patients but was discovered in several only by careful examination by experienced clinicians. It is probably fair to state that in ordinary practice the diagnosis of streptococcic sore throat would not have been made on clinical grounds for at least 25 per cent of this group of definitely infected patients. In the absence of special laboratory and bacteriologic study, these men would usually have been supposed to suffer from a "virus" type of infection of the respiratory tract.

Eleven patients in whom the maximum temperature was never more than 99 F. were of great importance. Generalized symptoms of an acute infection were almost completely lacking in these men. In military practice they would not have been removed from duty and in civilian life would not, in all probability, have consulted a physician. Definite clinical evidence of infection was present in 8, and in several the total leukocyte count or erythrocyte sedimentation rate was increased.

The serum antistreptolysin titer was measured at the onset of the acute illness and during convalescence. A significant increase in titer was frequently observed, even though the systemic reaction was slight or the physical signs usually associated with hemolytic streptococcus sore throat were minimal or absent. An antibody response did not occur after some of the most severe and typical infections.

The foregoing observations demonstrate that nearly all the persons who became carriers of the epidemic type of group A streptococci after exposure to infected food were infected and that the syndrome usually designated as follicular tonsillitis was frequently observed. There were, however, many mild and atypical cases in which the correct etiologic agent would not usually have been recognized in the absence of special laboratory study. It is probable that just such atypical diseases of the respiratory tract as these, occurring as the result of non-food-borne natural infection, have been responsible for the initiation of the many instances of rheumatic fever and other types of poststreptococcic nonsuppurative disorders in which a past history of infection by these organisms has not been obtained.

The complete suppression of tissue invasion by hemolytic streptococci must be the goal of all control measures for the prevention of late complications. Their eradication by technics for the treatment of the acute phase of the disease of the respiratory tract must fail, because many infected persons will not become sufficiently ill to require medical care and others will fail to present signs permitting the clinical recognition of the cause of the disease.

Suppurative and nonsuppurative complications, as sequelae to the disease caused by the epidemic streptococci, were found to be consistently absent, in contrast with the results of study of similar infections by a variety of other types during the same season.[6] This absence of complications is probably the result of the failure of the infecting strain to elaborate some essential toxin or other bacterial product. Similarly, this organism did not induce the formation of a rash in persons with positive Dick reactions or stimulate an antifibrinolysin response. In spite of these deficiencies, the strain was highly invasive, only 3 of the 35 men who became carriers having escaped infection as determined by clinical and immunologic observations.

A number of men previously infected by other types of group A streptococci were hospitalized at the time of the epidemic and were exposed to reinfection. Study of their cases revealed a curious phenomenon that deserves special consideration. None of these men reacted to the presence of the new type with more than minimal clinical signs of infection, yet in nearly one half significant increase in the total leukocyte count and/or erythrocyte sedimentation rate was demonstrated. Previous infection by another type had prevented the usual clinical signs of hemolytic streptococcus sore throat from developing but had not prevented a tissue reaction as the result of invasion of the throat by the epidemic type.

Evidence has been presented elsewhere [6] which strongly suggests that reinfection by new types of group A streptococci may be important in the pathogenesis of rheumatic fever and the other phenomena following streptococcic infections. The facts just stated suggest that such reinfection may often be unrecognizable clinically and may be demonstrable only by detailed bacteriologic study of the flora of the throat and by laboratory tests.

The results of this study show that the clinical signs and the course of hemolytic streptococcus disease of the respiratory tract may be highly variable and that the amount of tissue reaction and degree of antibody response is not well correlated with the severity of the illness. The nature of the disease is greatly altered by preceding infection by strains of another type.

It is hoped that these observations may focus attention on the large group of cases of mild and

atypical streptococcic infection and on their potential importance in the causation of rheumatic fever and other disorders now believed to be sequelae to infection by these organisms.

SUMMARY AND CONCLUSIONS

A large number of men simultaneously exposed to infection by group A streptococci were studied.

Tonsillectomy appeared to offer considerable protection against infection by the epidemic type.

The disease produced in different patients varied greatly in its severity and in the amount of tissue reaction demonstrated.

Many examples of mild or clinically atypical disease were observed which would not usually be recognized as instances of streptococcic infection.

Previous infection by another type of group A streptococcus profoundly modified the nature and course of the disease produced by the epidemic type.

SMALL ADENOMAS OF THE ADRENAL CORTEX IN HYPERTENSION AND DIABETES

CAPTAIN SIMON RUSSI AND CAPTAIN HERMAN T. BLUMENTHAL
MEDICAL CORPS, ARMY OF THE UNITED STATES
AND
LIEUTENANT COMMANDER SAMUEL H. GRAY, MC(S), U.S.N.R.

The principal observations generally reported concerning persons with tumors of the adrenal cortex consist of obesity, purple striae of the skin, acne, polycythemia, osteoporosis, imbalance of plasma electrolytes, diabetes, hypertension and genital dystrophy. These signs and symptoms have been observed in persons with adenomas and hyperplasias as well as in those with carcinomas of the adrenal cortex, and the total picture is usually referred to as Cushing's disease or adrenogenital syndrome. In general, even the benign tumors which produce this clinical entity occupy a large portion of the adrenal gland. There are, in addition, as Kepler and Keating [1] have pointed out, a large number of small cortical adenomas which are encountered incidentally in the course of routine autopsies in persons in whom there was no apparent clinical evidence of endocrine disease during life. The investigations presented here were undertaken in order to determine whether or not an examination of the clinical and pathologic data, in retrospect, would show the presence of certain endocrinologic disturbances not noted during the life of those patients. This report contains a survey of 131 cases of small adenomas of the adrenal cortex encountered in the course of 9,000 consecutive necropsies.

RESULTS

Incidence, Age and Sex Distribution.—The incidence of cortical adenomas in this series, 131 cases in 9,000 autopsies, is 1.45 per cent. However, since these necropsies were performed by many different persons, it is probable that there were many instances in which small tumors were missed or not recorded and that the actual frequency with which these tumors occur probably is slightly higher. Seventy-one of the adenomas (54.2 per cent) occurred in males, while 60 (45.8 per cent) were observed in females. Since the male to female ratio in our series is about 2:1, the incidence of adrenal adenomas in relation to sex is 2 per cent in females and 1.2 per cent in males.

TABLE 1.—*A Comparison of the Age Distribution in One Hundred and Thirty-One Cases of Adenomas of the Adrenal Cortex with That in Nine Thousand Autopsies*

Age by Decades	Age Distribution in 131 Cases of Cortical Adenoma		Age Distribution in 9,000 Consecutive Autopsies	
	Number	Per Cent	Number	Per Cent
Premature	0	0	270	3.0
0- 9	1	0.8	675	7.5
10-19	0	0	234	2.6
20-29	0	0	441	4.9
30-39	2	1.5	729	8.1
40-49	18	13.7	1,125	12.5
50-59	34	26.0	1,719	19.1
60-69	43	32.8	1,935	21.5
70-79	18	13.7	1,449	16.1
80-89	15	11.5	387	4.3
90-100	0	0	36	0.4
Total	131	100.0	9,000	100.0

A comparison of the age distribution in the 9,000 necropsies with that in the 131 cases of adenomas of the adrenal cortex is shown in table 1. It can be seen that only 3 patients with adenomas were found below the age of 40 years, after which there was a gradual rise in the incidence until a peak was reached in the decade between 60 and 69 years; this was followed by a decline in the next two decades. While these figures alone might imply a gradual rise in the incidence of these tumors, with a maximum being reached during the seventh decade, a comparison with the age distribution in autopsies in general shows that both have a similar curve.

The investigations were aided by a grant from the David May-Florence G. May Fund.

This article is based entirely on investigations conducted at Snodgras Laboratory, City Hospital and Laboratory of the Jewish Hospital in St. Louis.

This article has been released for publication by the Division of Publications of the Bureau of Medicine and Surgery of the United States Navy. The opinions and views set forth are those of the writers and are not to be construed as reflecting the policies of the Navy Department.

1. Kepler, E. J., and Keating, F. R., Jr.: Diseases of the Adrenal Glands: Tumors of the Adrenal Cortex, Diseases of the Adrenal Medulla and Allied Disturbances, Arch. Int. Med. **68**:1010-1036 (Nov.) 1941.

However, the possibility that there is such an increase in incidence with increasing age cannot be definitely eliminated by this comparison.

Anatomic Characteristics.—As just stated, we studied small adenomas which were incidentally found at necropsy. These varied from small microscopic nodules (3 instances) to tumors approximately 6 cm. in diameter (1 instance). The average size was about 1 cm. in diameter. Representative gross sections are shown in figure 1.

In 68 instances there was a record of the side on which the tumor was found. In 21 cases (31 per cent) it was found on the right side and in 34 cases (50 per cent) on the left side, while there were 13 instances (19 per cent) in which bilateral adenomas were observed. Multiple nodules in a single gland were found in only 2 specimens.

tissue adjacent to the tumor. As a result, the adenoma frequently appeared paler than the neighboring cortical tissue when the section was examined with the naked eye or the hand lens. The second type of cortical cell generally showed a centrally located nucleus more vesicular than that seen in the first type, and the cytoplasm was either homogeneously eosinophilic or contained finely dispersed lipid droplets. The relative numbers of each cell type varied in different tumors, but, in general, the first type predominated, although occasionally they were present in approximately equal numbers. In figure 2 there is shown a tumor in which the vacuolated type of cell was predominant, but islands of smaller solid cells were also present, and these are indicated by arrows.

Fig. 1.—Gross appearance of cortical adenomas on cross section. (Specimen on the extreme left is a cross section of a normal adrenal gland.)

Microscopically these nodules were sharply demarcated but not definitely encapsulated. In no instance was there any evidence of malignant invasion. The general morphologic structure of the individual cells was essentially similar to that of the cells forming the cords of the adrenal cortex. There was usually some distortion in the cellular outline, apparently due to the resistance of neighboring structures to the new growth. The cordlike arrangement of the cells had undergone varying degrees of distortion. In some areas the cords, although distinctly visible, appeared angulated, arched and of varying length. The cords failed to arrange themselves in parallel rows. In other areas the cord arrangement was entirely obliterated. The cortical cells composing the adenomas were of two types. The first of these contained either a centrally or eccentrically placed hyperchromatic nucleus and cytoplasm filled with medium-sized and large-sized lipid vacuoles. The lipid vacuoles were usually larger than those seen in the cells of the normal cortical

DISEASES ASSOCIATED WITH SMALL ADENOMAS OF THE ADRENAL CORTEX

A survey of the clinical data showed hypertension and diabetes to be the only diseases occurring with sufficient frequency to warrant an analysis of their relation to these small tumors of the adrenal cortex. Accordingly, we are presenting the data used to determine whether or not these diseases occurred with significantly greater frequency in persons with cortical adenomas than in the general group of autopsy.

1. *Hypertension.*—Both clinical and necropsy data were used in order to determine the presence of hypertension. The determination of clinical evidence of hypertension was patterned according to certain of the criteria used by Master, Marks and Dack,[2] whose data we have also

2. Master, A. M.; Marks, H. H., and Dack, S.: Hypertension in People over Forty, J. A. M. A. **121**: 1251-1256 (April 17) 1943.

used to compare with certain of our observations. Two divisions of the clinical group were made, (*a*) those patients with a blood pressure of 140 systolic and 90 diastolic or over and (*b*) those with a diastolic pressure of 95 or over. The pathologic evidence for the existence of hypertension was the presence of cardiac hypertrophy (weight of the heart in excess of 350 Gm.) in the absence of a valvular or myocardial lesion other than infarction due to coronary occlusion. No attempt was made to compare this evidence of hypertension with the sphygmomanometric data recorded in the clinical history of the 9,000 subjects studied by autopsy. Many of these were seen for the first time when they were dying, when the evaluation of the preexisting hypertension was extremely difficult. Instead, our autopsy data are compared with the data on living persons reported by Master, Marks and Dack and covering approximately 15,000 patients. This comparison is shown in table 2.

From the data of Master, Marks and Dack [2] it was determined that when the less critical criterion for the presence of hypertension was used (blood pressure 140 systolic and 90 diastolic), over half of the patients examined showed evidence of hypertension. On the other hand, when the more critical basis was employed (diastolic tension over 95), the percentage of those with hypertension fell to 22.4. The latter is more in agreement with the figure obtained by us on the basis of cardiac hypertrophy, namely 13.9 per cent. As to the percentages in various decades, it may be noted that there was definitely an increase after the age of 60 as compared with previous decades, although there were some minor fluctuations which were probably not significant but which were always above the percentages found in earlier decades.

As far as the 131 patients with cortical adenomas are concerned, there were 100 for whom complete clinical data were available for comparison with the necropsy observations. Seventy of these showed evidence of some degree of hypertension, while for 30 there were no clinical and pathologic evidences. In addition, there were 22 others in whom definite cardiac hypertrophy was present but for whom there were insufficient clinical data to determine the presence of a hypertensive state. Finally, there were 9 subjects who had to be eliminated because of valvular lesions which produced a cardiac hypertrophy and abnormality in blood pressure.

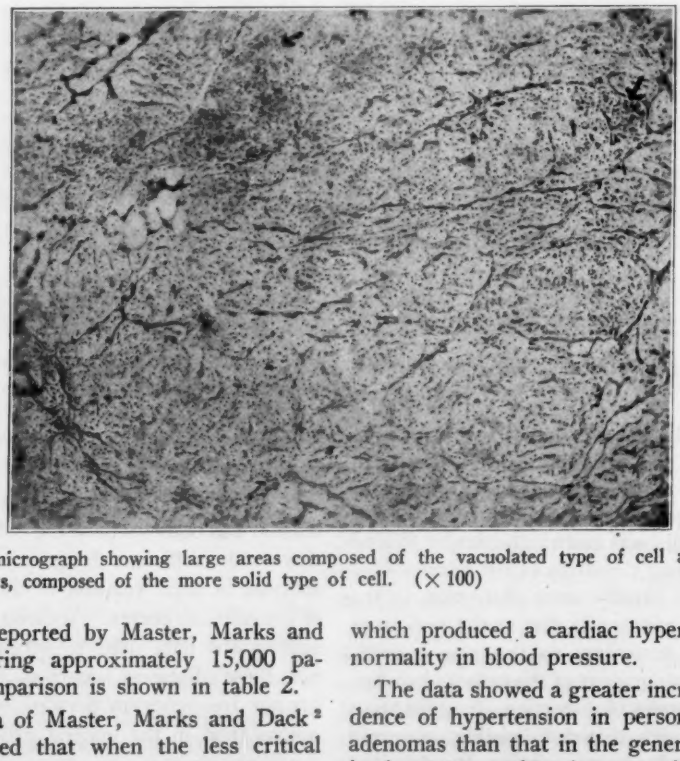


Fig. 2.—Photomicrograph showing large areas composed of the vacuolated type of cell and smaller areas, indicated by arrows, composed of the more solid type of cell. (× 100)

The data showed a greater increase in the incidence of hypertension in persons with adrenal adenomas than that in the general group examined at autopsy, by whatever criteria were used. If the less critical criterion (blood pressure 140 systolic and 90 diastolic) is used, there is an incidence of 70 per cent in the group with adenomas, as compared with 54.9 per cent in the clinical group presented by Master, Marks and Dack. If a diastolic pressure in excess of 95 is used as the basis, the group with cortical adenomas shows an incidence of 42 per cent, as compared with 22.4 per cent in the clinical group.

Finally, if a comparison is made on the basis of observations at autopsy, the group with adenomas shows an incidence of 72.9 per cent, as compared with 13.9 per cent in the general group examined at autopsy. It should be noted that in 96 per cent of the persons with adrenal adenomas having a blood pressure of 140 systolic and 90 diastolic or over there was evidence of cardiac hypertrophy at autopsy. A further breakdown of the data for the 100 persons with cortical adenomas according to the degree of hypertension shows that there were 70 with a blood pressure of 140 systolic and 90 diastolic or over, 57 with 150 systolic and 90 diastolic or over and 35 with a tension of 150 systolic and 100 diastolic or over. Of the 13 with a blood pressure between 140 systolic and 90 diastolic and 150 systolic and 90 diastolic, only 3 failed to show evidence of cardiac hypertrophy. As far as the percentages at various decades are concerned, the relation in the group with adrenal adenomas is essentially similar to that seen in the general clinical and autopsy groups. The apparent exception is in group 3 B (table 2), but this is probably not significant and may be due to the relatively small number of patients.

When the hypertensive persons are grouped according to decade (table 3), it can be seen that the greatest incidence in all groups generally lies between the ages of 40 and 69. More specifically, it may be noted that in the four groups comprising this study the maximum number is reached in the seventh decade of life in three of them and in the sixth decade in one. In the clinical group of Master, Marks and Dack, both subclassifications show a maximum between the ages of 40 and 49. While the incidence of hypertension is apparently higher after the age of 60 in all groups, including those with adenomas of the adrenal cortex, the greatest incidence in a given decade is reached before the age of 70.

Finally, in presenting the data on hypertension, we should emphasize that there were only 14 hypertensive persons among the 92 persons with adenomas of the adrenal cortex (15.2 per cent) in whom there was also a renal lesion to which the hypertension might possibly be attributed.

2. *Diabetes.*—In the analysis of the data it was observed that diabetes also occurred frequently in persons with cortical adenomas. It was noted that there were 270 persons with diabetes in the general group of 9,000 examined by autopsy, an incidence of 3 per cent. On the other hand, among the 131 persons with cortical adenomas there were 21 with proved diabetes, or an incidence of 16 per cent. Of these 21 persons, 16 were also in the group with both clinical and pathologic evidence of hypertension and 3 others in the group with cardiac hypertrophy without sufficient clinical evidence of an elevation in blood pressure, while only 2 were in the unclassified group. Thus, as in the case of hypertension, a comparison of the incidence of diabetes in the general autopsy group and in the group with adrenal adenomas shows an approxi-

TABLE 2.—*A Comparison of the Incidence of Hypertension in a Clinical Group with That in an Autopsy Group and a Group with Adenomas of the Adrenal Cortex*

Group	Number of Cases	Number of Hypertensive Persons	Percentage of Hypertensive Persons	Age of Hypertensive Persons (Percentage by Decade)†					
				0-39	40-49	50-59	60-69	70-79	80-100
1. Clinical group *									
a. Blood pressure 140/90 or over	14,849	8,148	54.9	....	36.6	55.3	71.4	79.2	81.5
b. Diastolic pressure 95 or over	14,849	3,320	22.4	....	11.6	21.9	27.7	33.4	30.7
2. Autopsy group									
Cardiac hypertrophy	9,000	1,254	13.9	4.1	17.5	16.2	23.0	19.7	22.6
3. Adrenal adenoma group									
a. Blood pressure 140/90 or over	100	70	70.0	50.0	42.9	68.0	78.8	91.8	64.3
b. Diastolic pressure 95 or over	100	42	42.0	0	42.9	52.0	42.4	41.7	28.6
c. Cardiac hypertrophy at autopsy	122	89	72.9	66.6	50.0	72.4	78.0	88.8	66.6

* The clinical group was obtained by combining the data for the two sexes in the article by Masters, Marks and Dack.[2]
† These figures were obtained by dividing the number of hypertensive persons in a given decade by the total number of persons (normal plus hypertensive) for that decade.

TABLE 3.—*A Comparison of the Age Distribution of the Hypertensive Persons in Various Groups*

	Percentage of the Total Number of Hypertensive Persons					
	0-39	40-49	50-59	60-69	70-79	80-100
1. Clinical group						
a. Blood pressure 140/90 or over	...	28.0	25.2	25.5	15.6	5.7
b. Diastolic pressure 95 or over	...	30.0	24.5	24.3	16.1	5.0
2. Autopsy group						
Cardiac hypertrophy.	6.9	14.1	19.8	31.8	20.4	7.0
3. Adrenal adenoma group						
a. Blood pressure 140/90 or over	1.4	8.6	24.3	37.1	15.7	12.9
b. Diastolic pressure 95 or over	...	17.1	31.4	28.6	14.3	8.6
c. Hypertrophy of heart	2.2	9.0	22.5	35.9	19.1	11.2

mately fivefold increase in the incidence in the latter over the former. In addition, in almost all instances in which diabetes was present in association with an adenoma evidence of hypertension was also present.

COMMENT

The results presented here show that the incidence of adenomas of the adrenal cortex was at least 1.45 per cent in 9,000 autopsies and that there is no significant difference in incidence between males (1.2 per cent) and females (2 per cent). There appears to be an increase in incidence up to the decade of 60 to 69 years and then a gradual decline, although it is possible that this is only a reflection of the age distribution in the autopsies as a whole.

Anatomically the tumors vary from small microscopic nodules to growths which reach a diameter of about 6 cm., the average being about 1 cm. in diameter. As stated, they are composed of two principal types of cells, the first with large lipid-containing vacuoles occupying the cytoplasmic area and the second with finely granular or solid cytoplasm. Ewing [3] has designated these small tumors as focal hyperplasias, in contradistinction to the larger tumors, which are true adenomas. According to him, the former may usually be distinguished from the latter in that the structure reproduces cortical zones whereas true adenomas have a typical neoplastic structure. The so-called true adenomas are believed to be relatively rare by Kelynack,[4] who found only 3 in 1,500 autopsies. As stated, the architectural pattern in a single tumor may vary from slight distortion of the cords to their complete obliteration. Thus there is probably no sharp distinction between focal hyperplasia and true adenoma on an anatomic basis.

Likewise, the functional distinction between small tumors of the adrenal cortex of the so-called clinically indifferent type and larger tumors which are associated with endocrinologic disorders is a relative one, since, as we have shown, even in association with the former hypertension and diabetes occur approximately five times as frequently as in the general autopsy group, and in many cases both diseases occur in the same person.

There exists considerable clinical and experimental evidence of the relation of the adrenal cortical hormones to hypertension and diabetes, but a careful evaluation of the clinical features is frequently difficult, particularly in the case of hypertension. There has often arisen some question as to the reliability of certain of the clinical criteria for the determination of the presence of this disease. Wilburne and Ceccolini [5] have recently presented several reasons for widely discrepant figures on the incidence of hypertension: namely, (1) a lack of agreement as to what blood pressure levels constitute arterial hypertension, (2) lack of rest periods during testing and (3) insufficient precautions for the elimination of such influences as dietary and psychic factors. Recently Master, Marks and Dack [2] stated that they consider the estimated incidence of hypertension in persons over 40 to be too high, and we have shown here that certain of their figures are considerably higher than those derived from observations at necropsy. Proof of hypertension by autopsy, as manifested by cardiac hypertrophy in the absence of valvular lesions, myocardial disease or congenital defects, offers the most reliable criterion when it can be employed.

Oppenheimer and Fishberg [6] have reviewed the literature concerning the association of adrenal cortical tumors with hypertension since it was first observed by Neusser,[7] in 1898, and have presented a number of additional cases. More recently Rinehart, Williams and Cappeller [8] have studied 26 cases of essential hypertension and have observed nodular or adenomatous hyperplasia in the adrenal cortex in a high percentage of these cases. On the other hand, Dempsey [9] has reported that in patients with essential hypertension the average weight of the adrenal gland was not significantly higher than in nonhypertensive control patients. He studied a total of 102 patients, of whom only 19 were classified as being definitely hypertensive. In all of this group there were only 9 with cortical adenomas, and of these only 3 were persons with essential hypertension. From this small number it was concluded that nodular or adenomatous hyperplasia of the adrenal cortex was not regularly found in association with essential

3. Ewing, J.: Neoplastic Diseases, Philadelphia, W. B. Saunders Company, 1941, pp. 830-831.

4. Kelynack, cited by Ewing.[3]

5. Wilburne, M., and Ceccolini, E. M.: A Note on the Incidence of Arterial Hypertension in 25,000 Army Examinees, Army M. Bull., 1943, no. 68, pp. 118-125.

6. Oppenheimer, B. S., and Fishberg, A. M.: The Association of Hypertension with Suprarenal Tumors, Arch. Int. Med. **34**:631-644 (Nov.) 1924.

7. Neusser, E., in Nothnagel, C. W. H.: Specielle Pathologie und Therapie, Vienna, A. Hölder, 1898, pp. 18 and 71.

8. Rinehart, J. F.; Williams, O. O., and Cappeller, W. S.: Adenomatous Hyperplasia of the Adrenal Cortex Associated with Essential Hypertension, Arch. Path. **32**:169-177 (Aug.) 1941.

9. Dempsey, W. S.: The Adrenal Cortex in Essential Hypertension, Arch. Path. **34**:1031-1034 (Dec.) 1942.

hypertension and that it occurred with considerable frequency in nonhypertensive persons. In general, each investigator has dealt with small numbers of cases, and the relation of these tumors to the total picture of the cause of hypertension could not be accurately evaluated. Recently Castleman and Smithwick [10] examined by biopsy the kidneys of 100 patients with clinical hypertension and observed that in more than half of them evidence of renal vascular disease was inadequate. The small number of patients with renal lesions in this group of persons with adrenal adenomas similarly indicates that hypertension cannot always be accounted for on the basis of pathologic changes in the kidneys. It should be noted further that Castleman and Smithwick observed that 6 of the 100 patients studied were found to have cortical adenomas. This incidence is in essential agreement with that of our much larger series, in which 89 of a total of 1,254 hypertensive patients (7.4 per cent) had this type of tumor. Additional evidence showing a relation between the adrenal cortical hormones and hypertension is found in the experiments of Helfrich, Cassels and Cole,[11] who observed that cortical extract is effective in preventing the usual fall in blood pressure when shock is produced, and in the observation by Loeb [12] that in 4 patients receiving desoxycorticosterone for the treatment of Addison's disease hypertension developed. In Loeb's cases it was not known whether or not these patients had underlying hypertensive disease masked by the Addison disease. The ability of desoxycorticosterone acetate to elevate the blood pressure to hypertensive levels is also demonstrated in the experiments of Rodbard and Freed.[13] These investigators showed that the injection of this substance into previously hypertensive dogs caused pulmonary edema to develop, while in previously nonhypertensive dogs an elevation in blood pressure was produced which persisted for variable periods after cessation of injections.

Certain experimental observations suggest the possibility that the adrenal hormones (1) may render a person more sensitive to renin or (2) that they may be instrumental in producing changes in the kidneys which would lead to an increased renin production. The former possibility is suggested by the experiments of Williams, Diaz, Burch and Harrison,[14] who found that adrenalectomized rats are noticeably less sensitive to renin than normal rats. The latter possibility is presented in the experiments of Selye and Hall.[15] These investigators observed that large doses of desoxycorticosterone acetate produced definite renal changes in fowl and dogs similar to those seen in human patients with nephrosclerosis. These changes were more readily obtained in fowl, but could also be produced in mammals, although it is probable that the refractory state is much longer in the latter than in birds. Opposed to these interpretations is our observation that in only 14 of the 92 patients with adrenal cortical tumors and evidence of hypertension (15.2 per cent) was there any degree of renal damage to which the hypertension might be attributed. It seems more likely that there is a direct effect of the cortical hormones on vascular tone, as indicated by the experiments on prevention of shock and that the renal damage observed by Selye and Hall [15] is due to a toxic effect of large doses of desoxycorticosterone.

The role of the adrenal hormones in the production of a hyperglycemic state has been amply reviewed by Britton and Silvette,[16] Soskin,[17] Kendall,[18] Evans [19] and Long, Katzin and Fry.[20] Suffice it to state here that it is now generally agreed that certain of the adrenal cortical hormones increase the rate of glyconeogenesis and

10. Castleman, B., and Smithwick, R. H.: The Relation of Vascular Disease to the Hypertensive State, Based on a Study of Renal Biopsies from One Hundred Hypertensive Patients, J. A. M. A. **121**:1256-1261 (April 17) 1943.

11. Helfrich, L. S.; Cassels, W. H., and Cole, W. H.: Cortical Extract in the Treatment of Shock: Preliminary Report, Am. J. Surg. **55**:410-426, 1942.

12. Loeb, R. F.: Adrenal Cortex Insufficiency, in Glandular Physiology and Therapy, Chicago, American Medical Association, 1942, pp. 287-305.

13. Rodbard, S., and Freed, S. C.: The Effect of Desoxycorticosterone Acetate on the Blood Pressure of the Dog, Endocrinology **30**:365-368, 1942.

14. Williams, J. R.; Diaz, J. T.; Burch, J. C., and Harrison, T. R.: The Relation of the Adrenal Glands to the Action of the Renal Pressor Substance, Am. J. M. Sc. **198**:212-219, 1939.

15. Selye, H., and Hall, C. E.: Pathologic Changes Induced in Various Species by Overdosage with Desoxycorticosterone, Arch. Path. **36**:19-31 (July) 1943.

16. Britton, S. W., and Silvette, H.: The Adrenal Cortex and Carbohydrate Metabolism, in Cold Spring Harbor Symposia on Quantitative Biology, Cold Spring Harbor, L. I., New York, The Biological Laboratory, 1937, vol. 5, pp. 357-359.

17. Soskin, S.: Metabolic Functions of the Endocrine Glands, in Luck, J. M.: Annual Review of Physiology, Stanford University, Calif., Annual Reviews, Inc., 1941, vol. 3, pp. 543-572.

18. Kendall, E. C.: The Function of the Adrenal Cortex, in Glandular Physiology and Therapy, Chicago, American Medical Association, 1942, pp. 273-286.

19. Evans, G.: The Adrenal Cortex and Endogenous Carbohydrate Formation, Am. J. Physiol. **114**:297-308, 1936.

20. Long, C. N. H.; Katzin, B., and Fry, E. G.: The Adrenal Cortex and Carbohydrate Metabolism, Endocrinology **26**:309-344, 1940.

by this means increase the blood sugar level. Clinically, it has been observed by Lukens, Flippin and Thigpen [21] that of 55 patients with carcinoma, hypernephroma, adenoma or hyperplasia of the adrenal cortex 27 (49 per cent) showed an impaired sugar tolerance compatible with a diagnosis of diabetes. The frequent presence of diabetes in association with cancers or large benign tumors of the adrenal cortex has already been mentioned.

In view of the fact that of the 21 patients with adrenal cortical adenomas and diabetes 19 also showed either clinical or pathologic evidence of hypertension, it is of interest to speculate whether these tumors elaborate a single hormone or produce the multiplicity of hormones in a manner similar to the normal adrenal cortex, but perhaps in excessive amounts. In the abundant literature on adrenal cortical hormones one finds evidence that desoxycorticosterone affects primarily salt and water metabolism, whereas corticosterone and its derivatives affect primarily sugar metabolism through glyconeogenesis, as already stated, and the amorphous fraction affects renal function. However, there may be some overlapping in these processes. Clinically, desoxycorticosterone is effective in raising the blood pressure of patients in shock, and it is also this substance which is suspected of producing hypertension in patients treated for Addison's disease, but it is relatively ineffective in raising the blood sugar level. This evidence would lead one to conclude that these tumors are probably able to elaborate more than one hormone and in this respect function in a manner similar to the normal adrenal cortex.

There remains, finally, to discuss the physiologic roles of the two types of cells noted in histologic sections. In 1942, in underfeeding experiments on guinea pigs, it was pointed out by Blumenthal and Loeb [22] that the degree of mitotic activity in the adrenal cortex is in inverse ratio to the number of lipid vacuoles present in the fasciculate cells. This principle has been observed to hold true by Blumenthal in a more extensive series of as yet unpublished experiments on guinea pigs. It has also been noted by Dosne and Dalton,[23] as well as by Selye,[24] that the amount of cortical lipid, as demonstrated by either a sudan stain or osmium tetroxide, definitely decreases as the adrenal gland enlarges with increased activity. More recently Sarason [25] has observed a decrease in cortical lipids in human beings with overwhelming infection and cachexia, as well as in rats under certain experimental conditions. In a second paper the same investigator [26] again observed a cortical enlargement associated with decreased amount of lipid in inflammatory diseases, cachexia, pemphigus and protracted emesis. This evidence suggests that under these conditions there occurs a discharge of lipid into the circulation and the cytoplasm of cortical cells changes from the vacuolated to the more solid or granular state. However, Sarason was unable to explain the observation that in hypertension there was a cortical enlargement associated with an increased amount of lipid.

It has recently been shown by Sayers, Sayers, White and Long [27] that 2 mg. of pituitary adrenotropic substance produces a distinct lowering of the adrenal cholesterol level in rate three hours after injection, while repeated injections over a period of three days result in an increase in adrenal cholesterol concentration above that of control animals. This experiment may be interpreted as showing that the immediate effect of a stimulus on the adrenal cortex is to release adrenal cortical hormones into the circulation, and it may reasonably be expected that this would be manifested histologically in the nonvacuolated, relatively solid type of cortical cell. Subsequent injections of pituitary adrenotropic substance may result in a condition in which the process of storage of hormones exceeds the output, resulting in an increase in adrenal cholesterol concentration. If subsequent stimuli continue to be strong, as for example in severe cachexia, the solid type of adrenal cell may remain predominant, because the adrenal hormones are probably being discharged as rapidly as they are formed. In interpreting the preponderance of vacuolated cells in the adenomas associated with hypertension, we suggest that there may be a

21. Lukens, F. D. W.; Flippin, H. F., and Thigpen, F. M.: Adrenal Cortical Adenoma with Absence of the Opposite Adrenal: Report of a Case with Operation and Autopsy, Am. J. M. Sc. **193**:812-830, 1937.

22. Blumenthal, H. T., and Loeb, L.: Two Antagonistic Effects of Underfeeding on Adrenal Cortex of the Guinea Pig, Am. J. Path. **18**:615-631, 1942.

23. Dosne, C., and Dalton, A. J.: Changes in Lipoid Content of Adrenal Gland of Rat Under Conditions of Activity and Rest, Anat. Rec. **80**:211-217, 1941.

24. Selye, H.: Studies on Adaptation, Endocrinology **21**:169-188, 1937.

25. Sarason, E. L.: Morphologic Changes in the Rat's Adrenal Cortex Under Various Experimental Conditions, Arch. Path. **35**:373-390 (March) 1943.

26. Sarason, E. L.: Adrenal Cortex in Systemic Disease, Arch. Int. Med. **71**:702-712 (May) 1943.

27. Sayers, G.; Sayers, M. A.; White, A., and Long, C. N. H.: Effects of Pituitary Adrenotropic Hormone on Cholesterol Content of Rat Adrenal Glands, Pro. Soc. Exper. Biol. & Med. **52**:200-202, 1943.

relatively weak but persistent stimulus similar to repeated small injections of pituitary extract in the experimental situation and that, while the storage of lipids exceeds the rate of discharge, the latter may still be greater than that in the normal adrenal cortex. The presence of islets of solid cells in practically every cortical adenoma probably indicates the sites in which there is an active discharge of hormones at the same time that the process of storage is going on in other areas.

SUMMARY

The incidence of benign adenomas of the adrenal cortex in 9,000 routine autopsies was found to be at least 1.45 per cent. The incidence was 2 per cent in females and 1.2 per cent in males. There appeared to be a gradual increase in age incidence up to the seventh decade, although this may have been only a reflection of the age distribution of the autopsies as a whole.

Hypertension and diabetes occurred five times as frequently in persons with cortical adenomas as in the general autopsy group, and both diseases were frequently present in association with such a tumor in a single person.

The relation of the adrenal hormones to diabetes and hypertension was studied, and hypotheses concerning the possible role of the vacuoles and solid cells comprising the tumors in the production and storage of these hormones were formed.

PAROXYSMAL HEMOGLOBINURIA DUE TO THE COLD HEMOLYSIN

OBSERVATIONS WITH A REPORT OF A CASE OF ITS OCCURRENCE IN AN AERIAL GUNNER

LIEUTENANT COLONEL EDWARD D. MAIRE
MEDICAL CORPS, ARMY OF THE UNITED STATES

Hemoglobinemia with resultant hemoglobinuria is exhibited in several varied clinical conditions. Notably these are (*a*) in a certain percentage of burns; (*b*) in some severe infectious diseases, including the exanthemas; (*c*) after extended physical exertion, as is seen in the so-called march hemoglobinuria[1] of the soldier, and probably more specifically a myoglobinuria; (*d*) from organic toxins, such as venom, the deadly mushrooms and favism (a sensitivity reaction)—a disease which might appear in troops in Italy, especially southern Sicily, where the bean Vicia faba is plentiful; (*e*) in poisoning with chemicals such as toluylene diamine, saponin and photodynamic fluorescent hydrocarbons, including sulfonamide drugs and acridine dyes;[2] (*f*) in blackwater fever sometimes complicating malaria; (*g*) in some forms of chronic hemolytic anemia known as Marchiafavia-Micheli, or nocturnal, hemoglobinuria[3]; (*h*) in severe cardiac decompensation; (*i*) from incompatibility in blood transfusions; (*j*) in reabsorption from internal hemorrhage, and (*k*) in certain cases of probable syphilis which develop hemoglobinuria in response to the formation and action of a cold hemolysin. Possibly the phenomenon is present also in small degree physiologically, although when hemolysis occurs slowly and is not too great the hemoglobin is readily converted to bilirubin, from which the liver then produces urobilin.[4] Also, up to a certain amount, hemoglobin passed through the glomeruli is taken up and conserved by the epithelium of the convoluted tubules.[5] A series of experiments for the study of hemoglobinuria[6] produced in dogs would indicate that the renal threshold for dog hemoglobin varies greatly, with an average of about 155 mg. per kilogram of body weight. This threshold was depressed by as much as 46 per cent on daily diminishing injections. All types may be paroxysmal in nature, but, generally speaking, it would be well to limit the title of paroxysmal to the nocturnal and cold hemolysin phenomena. Inasmuch as many troops are exposed to low temperatures (Arctic regions and high altitude flying), it is believed that the presentation of the pertinent findings for a patient suffering from the cold type of paroxysmal hemoglobinuria together with a review of some of the literature relative to the subject might be of value.

The first published report of a case of paroxysmal hemoglobinuria was that of a 10 year old child with congenital syphilis presented by Dressler in 1854.[7] In this case, as well as in a similar one published by Elliotson in 1831 and in another by Stewart in 1794,[7] the condition was cured with quinine, so it may have been caused by malaria. In 1866 Parry differentiated plain paroxysmal

1. Best, C. H., and Taylor, N. B.: The Physiological Basis of Medical Practice, ed. 2, Baltimore, William Wood & Company, 1939, p. 79.

2. Hueper, W. C.: Reactions in the Blood and Organs of Dogs on Intravenous Injections of a Solution of Hemoglobin, J. Lab. & Clin. Med. **29**:628 (June) 1944.

3. (*a*) Ham, T. H.: Studies on the Destruction of Red Blood Cells: I. Chronic Hemolytic Anemia with Paroxysmal Nocturnal Hemoglobinuria; an Investigation of the Mechanism of Hemolysis, with Observations on Five Cases, Arch. Int. Med. **64**:1271 (Dec.) 1939. (*b*) Ham, T. H., and Dingle, J. H.: Studies on the Destruction of Red Blood Cells: II. Chronic Hemolytic Anemia with Paroxysmal Nocturnal Hemoglobinuria; Certain Immunological Aspects of the Hemolytic Mechanism with Special Reference to Serum Complement, J. Clin. Investigation **18**:657 (Nov.) 1939.

4. Jones, C. M.: Study of Bile Pigments by Means of Duodenal Tube in Paroxysmal Hemoglobinuria. M. Clin. North America **5**:1421 (March) 1922.

5. Lichty, J. A., Jr.; Havill, W. H., and Whipple, G. H.: Renal Thresholds for Hemoglobin in Dogs. J. Exper. Med. **55**:603 (April) 1932. Havill, W. H.; Lichty, J. A., Jr.; Taylor, G. B., and Whipple, G. H.: Renal Threshold for Hemoglobin in Dogs Uninfluenced by Mercury Poisoning, ibid. **55**:617 (April) 1932. Havill, W. H.; Lichty, J. A., Jr., and Whipple, G. H.: Tolerance for Mercury Poisoning Increased by Frequent Hemoglobin Injections, ibid. **55**:627 (April) 1932. Newman, W. V., and Whipple, G. H.: Hemoglobin Injections and Conservation of Pigment by Kidney, Liver and Spleen, ibid. **55**:637 (April) 1932.

6. Gilligan, D. R.; Altschule, M. D., and Katersky, E. M.: Studies of Hemoglobinemia and Hemoglobinuria Produced in Man by Intravenous Injection of Hemoglobin Solutions, J. Clin. Investigation **20**:177 (March) 1941. Lichty and others.[5]

7. Mackenzie, G. M.: Paroxysmal Hemoglobinuria: A Review, Medicine **8**:159 (May) 1929.

hemoglobinuria from malarious blackwater fever. The same year Gull showed the relation of chilling to the phenomenon.[7] In 1880 Rosenbach originated the contemporary Rosenbach test, producing hemoglobinuria in susceptible persons by immersing the feet in iced water and then warming them. In 1881 Ehrlich demonstrated, by detecting free hemoglobin in the blood drawn from a finger to which a tourniquet had been applied at its base and then immersed in cold water, that hemoglobin was formed locally in the part chilled, probably due to the production of a lysin by the endothelial cells of the surface blood vessels. He thus agreed with Lichtheim (1876) who proved that hemoglobinuria is a result of excretion by the kidneys of hemoglobin that had been formed in the blood vessels and not, as had been previously believed, produced by the kidney.[7] In 1904 Donath and Landsteiner [8] made thorough serologic investigations of the phenomenon and made clear the following facts: (1) that a lysin is present in the plasma or serum of patients suffering from this disease which can unite with red blood cells only at low temperatures; (2) that the hemolytic action takes place in two phases: (*a*) the union of the lysin with the red cells at a low temperature and (*b*) lysis of the red cells on warming; (3) that complement is essential for the second phase of the reaction; (4) that the lysin is an isohemolysin and an autohemolysin, and (5) that the patient's red cells are not hemolyzed by the serum of a normal person of the same grouping (AB, B, A, O). These dictums of Donath and Landsteiner, shown diagrammatically in figure 1, are acceptable today.

Most of the earlier workers realized the association of syphilis, preponderantly of the congenital type, with the disease, there being syphilitic stigmas exhibited in about one third of the cases. Since the discovery of the Wassermann reaction (1906), proof of this association has become overwhelming. In a review by Donath and Landsteiner,[9] there were 95 persons with syphilis in 99 patients reported on. It is apparent that there may be two almost specific tissue reactions in infections with the Treponema pallidum, one producing the "reagin" of the Wassermann reaction and the other the "cold hemolysin." In 1925 Nanba [10] produced a cold hemolysin in rabbits by intraperitoneal injections of emulsions of organs (liver, kidney and spleen) of various animals. It had been shown that similar treatment will produce a positive reaction to the Wassermann test in rabbits. Clinically, however, production of the "cold hemolysin" in syphilis is rare. Whereas it can be said that 95 per cent of the persons with paroxysmal hemoglobinuria due to the cold hemolysin are also syphilitic, only a small portion of persons with syphilis exhibit paroxysmal hemoglobinuria. From the practical view, the decided value of antisyphilitic therapy as a cure of the disease is somewhat convincing evidence of its causation. Certainly it is a good guide for the clinician's approach to the problem. It is to be emphasized that this important place of syphilis applies in only the cases of the disease due to the cold hemolysin and not to any of the other forms mentioned in the opening paragraph.

While the serologic phenomena are qualitatively uniform, the signs and symptoms of the disease may vary considerably. Hemoglobinuria may develop readily in some susceptible persons. Thus Mackenzie [7] reported 1 patient suffering

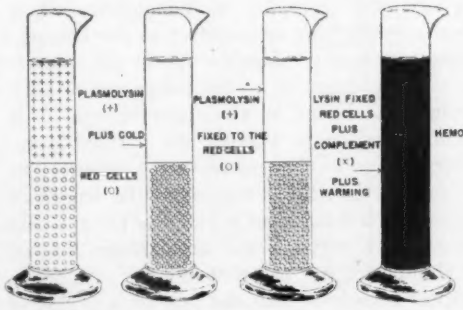


Fig. 1.—Mechanism of the cold hemolysin. The lysin (+) in the plasma is fixed to the red cells (O) by lowered temperature, the necessary degree of which depends on the individual susceptibility. Complement (×) plus rewarming produces hemolysis of the lysin-fixed cells.

a paroxysm in New York city during July. In other cases, as in the one reported in this paper, rather extreme low temperatures were required. The reaction, both the fixation of the lysin to the erythrocytes on exposure to the cold and the hemolysis of the red cells on warming, probably takes place in the surface vessels of the skin only. The inhalation of extremely cold air or the drinking of ice water will not precipitate an attack even in especially susceptible persons (Boas and Luzzati and Sorgente, cited by Mackenzie), whereas placing the hands or feet in ice water and then warming them, as in the Rosenbach test, may initiate a violent paroxysm. Stevenson [11]

8. Donath, J., and Landsteiner, K.: Ueber paroxysmale Hämoglobinurie, München. med. Wchnschr. **51:** 1590, 1904. Mackenzie.[7]

9. Donath, J., and Landsteiner, K.: Ueber Kälte Hämoglobinurie, Ergebn. d. Hyg., Bakt. **7:**184, 1925.

10. Nanba, M.: Ueber die künstliche Erzeugung des Autohämolysins, Deutsche med. Wchnschr. **51:**594 (April 10) 1925: cited by Mackenzie.[7]

11. Stevenson, I. P.: Paroxysmal Hemoglobinuria with Report of a Case, McGill M. J. **12:**192 (Oct.) 1943.

reported a case in which the ordinary Rosenbach procedure precipitated an almost complete anuria and the patient was critically ill for several days until renal function was gradually restored. Such a reaction could well be due to casts of hematin obstructing the glomerulotubular apparatus. Aside from such extremes are the persons with latent disease in whom, usually persons with syphilis, no signs or symptoms exist but the Donath-Landsteiner reaction of the blood is positive. Also, there are subjects in whom chilling will produce an albuminuria, general malaise, anemia and a leukocytosis but no hemoglobinuria. Thus, even though hemoglobinuria is not demonstrated clinically, in persons with unexplained anemia and transitory albuminuria the disease should be suspected. This is especially true if the patient is known to have syphilis. Less commonly does the individual patient's susceptibility vary, although it has been shown that the thermolability of the isolysin may vary from time to time in a given patient. Yorke and Macfie [12] demonstrated that after frequent episodes of hemoglobinuria a patient's serum may be negative to the Donath-Landsteiner test, presumably due to the exhaustion of complement or to the formation of anticomplement. From in vivo experiments, it is difficult to conclude whether the variety of responses is due to changes in the complement or the lysin. Also, the influence of the length of time to which the patient is subjected to the cold stimulus may vary. Yorke and Macfie showed that chilling for five to seven minutes resulted in a greater hemolysis than did that for a period of thirty minutes. They suggested that there might be some degradation of the lysin after long exposure to cold or that the longer time allowed a greater distribution of complement midpiece to more erythrocytes. No entirely satisfactory explanation can be given for the symptoms presented by various sufferers, although it is probable that the titer of the lysin determines the severity of the disease. The patient here reported on had no symptoms whatever during his attacks, even when, as in one paroxysm, the red cells fell to 2,510,000 and the hemoglobin content to 52 per cent. In other cases general malaise, chills and weakness have developed. Some patients exhibit pronounced vasomotor changes, such as a Raynaud syndrome, urticaria and vesicle formation in the epidermis. Albuminuria is a constant sign, and leukocytes and casts of pigment bodies are frequently found in the urine. At times the blood pressure falls, and at others it may become elevated. Often the patient's temperature increases.[7]

The case here reported is that of a 26 year old white man just finishing training as an aerial gunner with a heavy bomber crew. He had always been healthy and, indeed, had no untoward symptoms on presenting himself to the medical officer. He stated that his mother suffered from a mild form of arthritis but had always been well otherwise. The condition of his father, aged 53, and separated from the family, was unknown. One half-brother died from suffocation in infancy. There was no history of familial disease. The patient had spent six weeks in Hawaii with the navy in 1932 but had had no illness during that time. He had been married for six years, and his wife was well except for an indefinite cardiac complaint associated with fainting spells. She had had one pregnancy, which resulted in a spontaneous abortion in the early weeks "due to a fall." On induction into the army and on at least one routine check-up since, the reaction to the Kahn test of the blood had been reported negative. He had been proficient in his training, most of which had been at comparatively low altitudes, although temperatures as low as 21 F. (—5 C.) had been experienced without difficulty. Two weeks before complaining to his flight surgeon, the patient had noticed, soon after landing from a high altitude flight during which temperatures as low as minus 6 F. (—14 C.) had been reached, that the voided urine was black. Inasmuch as he felt completely well and as another specimen of urine voided a little later was of normal color, the patient dismissed the incident. He said that he had had moderate difficulty with the oxygen mask frosting on the flight, and he felt that this had been the cause. He had been chilly while at the high altitude, in spite of the sheepskin-lined clothes worn. He made similar flights on ten succeeding nights, and each time he voided one specimen of black urine three or four hours after landing. On one occasion he noted that the urine passed immediately after landing was of normal color but urine voided three hours later was black. On study, the patient appeared rugged and well. The physical examination revealed a soft functional systolic murmur at the base of the heart, normal blood pressure readings and active pupillary, cutaneous and deep tendon reflexes. The reaction of the blood to the Kahn test was negative, but a Wassermann test asked for after this finding was reported as eliciting a positive reaction. The red cell count was 3,580,000, with a hemoglobin content (Sahli) of 78 per cent and a leukocyte count of 6,300, normally distributed differentially. The sedimentation rate was 20 mm. in one hour. These values were about the same on three successive examinations within the next four days. The fragility of the red cells to saline solution showed hemolysis beginning at 0.42 per cent and complete at 0.36 per cent (within normal limits). The nonprotein nitrogen content of the blood was 30, and the sugar content was 93 mg. per hundred cubic centimeters. The reaction to the Donath-Landsteiner test was positive, there being rather pronounced hemolysis of both the cells of the patient and the cells from a normal person of a similar group *(A)* by the patient's serum. The serum of the normal control did not hemolyze the red cells of the patient. Results of tests for isoagglutinins were negative. The icteric index was 5. (Of special interest was the demonstration once again of the fact that the lysin is in the susceptible person's serum and that this lysin leaves the serum to become fixed in the red cells when exposed to the cold. Here, a 2 per cent saline suspension of the patient's washed red cells was

12. Yorke, W., and Macfie, J. W. S.: The Mechanism of Autolysis in Paroxysmal Haemoglobinuria, Brit. J. Exper. Path. **2**:115 (June) 1921.

mixed with an equal volume of his serum and refrigerated for ten minutes at 4 C. Immediately after refrigeration the serum was drawn off and was used to perform a complete Donath-Landsteiner test, only the red cells of a normal person being used. No hemolysis took place.) The urine was clear and yellow, with a specific gravity of 1.007 and a p_H of 6.5. It contained no glucose but did show a trace of albumin and an occasional leukocyte. It is of interest to note that the acidity of this specimen of urine associated with a trace of albumin and some leukocytes in the sediment because, as will be seen, during a paroxysm induced later the urine became strongly acid, the albumin increased and blood cells and pigment casts appeared (table 1). On the succeeding days the urine was entirely normal and albumin free, and the p_H varied from 6.5 to 7.5. At first, the patient was observed after he had sat on the open porch for thirty minutes with the temperature at 32 F. (0 C.). Even though clothed in light summer underwear and trousers and a light woolen shirt, he did not become uncomfortable during the period nor did hemoglobinuria develop. He was then placed in a large storage refrigerator for one hour at 28 F. (—2 C.), and again he had no untoward symptoms. A few days later he was exposed to the same temperature for one hour, followed by a temperature of 21 F. (—6 C.) for forty-five minutes. During the latter exposure the patient complained of being chilly—"about the same as at high altitude." Blood counts and urinalyses before and after this chilling, demonstrating a paroxysm of hemoglobinuria, are recorded in table 1 *A*. Photographs of the urine are shown in figures 2 and 3. After three days the erythrocyte count had returned to 3,640,000, the hemoglobin content to 70 per cent and the leukocyte count to 7,400, with polymorphonuclear cells 59 per cent, lymphocytes 30 per cent, mononuclears 12 per cent, eosinophils 5 per cent and myelocytes 3 per cent. At the end of twenty days the red cells had returned to 4,360,000, with 70 per cent hemoglobin. The sharp increase in the acidity of the urine to a p_H of 5.5 was striking and worthy of discussion.

TABLE 1.—*Comparison of Condition of Patient During Refrigeration Experiments*

	Blood			Urine				
Elapsed Time	Red Blood Cells	White Blood Cells	Hemoglobin, %	Appearance	Specific Gravity	Albumin	Sugar	Microscopic	p_H
				A					
Before refrigeration....	3,710,000	7,800	72	Clear, yellow	1.014	0	0	Negative	6.0
After refrigeration zero	3,680,000	4,950	70	Cloudy, straw	1.012	0	0	Few white cells	6.0
½ hour	3,700,000	5,100	75	Cloudy, straw	0.012	0	0	Few white cells	6.0
1 hour	3,715,000	4,800	72	.....	.....	..	..	.....	...
1½ hours	3,500,000	5,500	70	.....	.....	..	..	.....	...
2 hours	3,420,000	6,200	70	Cloudy, straw	0.014	0	0	Few white cells	6.0
2½ hours	3,200,000	7,600	65	Cloudy, straw	1.010	Trace	0	Many white cells	5.5
3 hours	3,000,000	9,400	62	Brown	1.021	1+	0	Many white and red cells and pigment casts	5.5
3½ hours	2,510,000	13,500	52	Very dark	1.022	1+	0	Many white and red cells and pigment casts	5.5
4 hours	2,600,000	11,000	55	Slightly dark	1.020	1+	0	Many white and red cells and pigment casts	5.5
5 hours	.....	.....	..	Clear, straw	1.010	0	0	Few white cells	7.0
				B					
Before refrigeration....	3,830,000	6,300	70	Clear, yellow	1.012	0	0	Negative	7.5
After refrigeration zero	3,800,000	5,100	70	Clear, yellow	1.014	0	0	Negative	7.5
½ hour	3,560,000	4,800	65	Clear, yellow	1.012	0	0	Negative	7.5
1 hour	3,750,000	5,100	68	Clear, yellow	1.018	0	0	Negative	7.5
1½ hours	3,780,000	6,400	70	Clear, yellow	1.014	0	0	Negative	7.5
2 hours	4,040,000	7,100	70	Clear, yellow	1.008	0	0	Negative	6.5
2½ hours	4,260,000	8,300	72	Clear, straw	1.006	0	0	Negative	6.5
3 hours	4,180,000	7,600	72	Clear, yellow	1.010	0	0	Negative	7.5
3½ hours	4,010,000	6,600	70	Clear, yellow	1.012	0	0	Negative	7.5
4 hours	4,260,000	5,800	72	Clear, yellow	1.012	0	0	Negative	7.5
5 hours	4,100,000	4,900	70	Clear, yellow	1.010	0	0	Negative	7.5
				C					
Before refrigeration....	4,520,000	4,800	76	Clear, yellow	1.010	0	0	Negative	6.0
After refrigeration zero	4,600,000	4,500	75	Clear, yellow	1.010	0	0	Negative	6.0
½ hour	4,200,000	6,200	72	Clear, yellow	1.010	0	0	Negative	6.0
1 hour	4,120,000	6,000	70	Cloudy, straw	1.016	0	0	Few white cells	6.0
1½ hours	3,800,000	6,500	68	Cloudy, straw	1.020	0	0	Few white cells	6.0
2 hours	3,810,000	7,800	62	.....	.....	..	..	.....	...
2½ hours	3,600,000	9,000	60	Cloudy, straw	1.020	2+	0	Many white and red cells	5.5
3 hours	3,400,000	11,000	58	Cloudy, brown	1.022	2+	0	Many white and red cells	6.0
3½ hours	4,000,000	9,200	60	Cloudy, yellow	1.018	Trace	0	Many white cells	7.0
4 hours	4,120,000	8,700	66	Clear, straw	1.010	0	0	Negative	7.0

In *A* are tabulated the blood and urinary findings during the refrigeration experiment on the patient's admission and without medication, while in *B* are the findings in the same experiment after rapid and pronounced alkalization of the urine with sodium bicarbonate has given the patient protection. In *C* the patient is still receiving alkali, but the efficaciousness has diminished. The latent period following the chilling and warming is well demonstrated in *A* and *C*. See table 2 for Donath-Landsteiner tests corresponding to these experiments.

In his studies of the destruction of the red blood cells in cases of paroxysmal nocturnal hemoglobinuria (Marchiafava-Micheli type), Ham [3a] surmised that the hemoglobinuria might be due to a decrease of the p_H of the blood, it having been demonstrated that the carbon dioxide content of the blood was increased during the hypoventilation of the lungs during sleep.

To combat this "acidity," Ham administered 65 Gm. of sodium bicarbonate during the twenty-four hours prior to testing and found a pronounced reduction of hemoglobinuria in 2 cases. It has also been observed that thorough alkalization of the urine is of benefit in combating the hemoglobinuria of blackwater fever.[13] There are no reasons to believe that the carbon dioxide content of the blood in the patient here reported was increased either during his flights at high altitude, when oxygen was used continuously above 10,000 feet (3,000 meters), or during the refrigerator

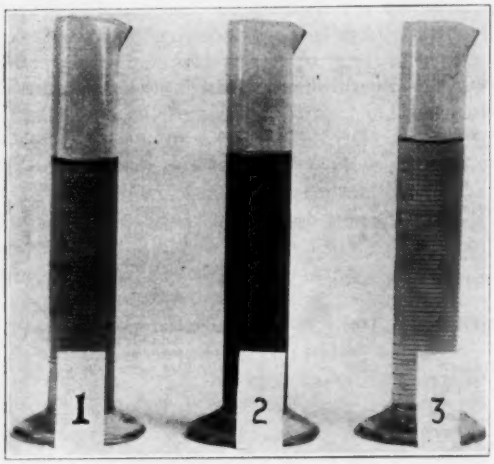


Fig. 2.—The graduate marked 1 is a photograph of the patient's urine prior to exposure to the cold; graduate 2 illustrates the intensity of the hemoglobinuria occurring three hours after the exposure was stopped, and graduate 3 demonstrates the clear urine another two hours later.

experiments. Nevertheless, the urine became intensely acid. It was this observation that prompted the decision to "alkalize" the patient and to repeat the experiment as shown in *B* of table 1. It is seen that after the administration of 32 Gm. of sodium bicarbonate in twenty hours the urine reached a p_H of 7.5, the Donath-Landsteiner test was less positive and that hemoglobinuria with the associated blood changes did not develop during the exact reduplication of the refrigeration and warming. However, a repetition of the same experiment after the patient had been given 12 Gm. of the alkali daily for a period of thirty days demonstrated a relapse in spite of an alkaline urine. During this period several tests revealed a positive Donath-Landsteiner reaction. In *C* of table 1 are recorded the phenomena of the "cold" hemolysin once more at work. In table 2 are shown the results of the Donath-Landsteiner tests done with each experiment *A*, *B* and *C* of table 1. In *D* of table 2 is shown an isolated test done on rapid, intensive alkalization of the urine with sodium bicarbonate for the first time. As the results of this test were entirely negative, it would appear that the drug was most efficacious when first administered, losing its effectiveness by degrees as time went on.

Several authors have studied the mechanism of hemoglobinuria and the effects of hydrogen ion concentration on it.[14] It is probable that a vasodilatation of the capillaries[14a] allows the large hemoglobin molecule (molecular weight 68,000) to pass through the renal glomerulus and into the tubule, where some reabsorption takes place.[6] The process of reabsorption may depend on one

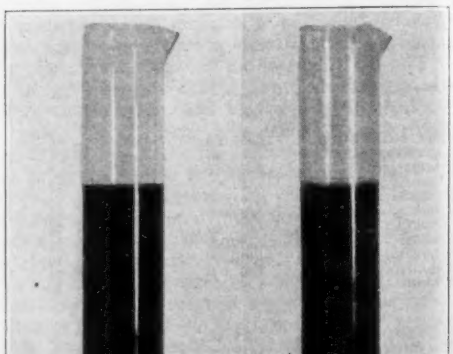


Fig. 3.—The graduate marked 1 illustrates the hemoglobinuria two hours after flight, while graduate 2 shows the urine three hours following exposure to the cold in the refrigeration experiment. The hemoglobinuria following the patient's flight is more intense.

or both of two factors: (*a*) the electrical charge carried by the protein molecule itself and (*b*) the charge carried by the tubule membrane, the electrical charge associated with the various

13. (*a*) Baker, S. L., and Dodds, E. C.: Obstruction of Renal Tubules During Excretion of Hemoglobin, Brit. J. Exper. Path. **6**:247 (Oct.) 1925. (*b*) Strong, R. P.: Stitt's Diagnosis, Prevention and Treatment of Tropical Diseases, ed. 6, Philadelphia, The Blakiston Company, 1942, vol. 1, p. 160. (*c*) Circular Letter no. 56, United States War Department, Office of the Surgeon General, Washington, D. C., Government Printing Office, 1942.

14. Yuile, C. L.: Hemoglobinuria, Physiol. Rev. **22**:19 (Jan.) 1942.

14a. Krogh, A.: The Anatomy and Physiology of Capillaries, New Haven, Conn., Yale University Press, 1929.

hydrogen ion concentrations effecting changes in the permeability; alkalinity favoring and acidity tending to prevent reabsorption.[15] Webster and colleagues[16] demonstrated, in experiments on frog kidney perfused with hemoglobin solutions of various p_H, that acidity increased and alkalinity decreased the elimination of hemoglobin. They suggested that the more acid solutions increased the permeability of the glomerular capillaries, letting more molecules of hemoglobin through, or that the lower p_H values changed the dissociation state of the molecule, allowing it to pass the negatively charged glomerular membranes more easily. Walbum[17] showed the effects of hydrogen ion concentrations on the hemolyzing properties of various bacteriolysins (Bacillus welchi, Clostridium septicum, Bacillus oedematiens and Bacillus tetani), using in vitro experiments on horse blood corpuscles. He demonstrated an optimum and a minimum point for hemolysis, below and above which p_H the degree of hemolysis would be increased or decreased respectively. The hemolytic action varied for each bacteriolysin. His work deserves greater investigation, with possibilities of valuable clinical application in therapy. Baker and Dodds[13a] showed that when the concentration of sodium chloride is 1 per cent urine with a p_H of below 6.0 will precipitate hemoglobin in the kidney tubule, possibly causing obstruction. They showed that in vitro hemoglobin is thrown out of solution when the p_H of the medium is not more than 6.0 and the sodium chloride content 1 per cent or more. They suggest alkalization of the urine in all forms of hemoglobinuria—blackwater fever, transfusion reactions and paroxysmal hemoglobinuria. De Navasquez[18] disagreed with the theory of precipitation of hemoglobin in the tubules when the urine is more acid. He reported 2 cases of death following transfusion reactions in which no such intratubular precipitation occurred in spite of an acid urine. He stated that hemoglobin is excreted in both alkaline and acid urines provided the volume flow through the glomerulus is sufficient; in fact, he demonstrated a better elimination of hemoglobin and its products with acid urine when the volume flow was the same. In the case here presented it is suggested (in *D* of table 2) that the p_H of the kidney urine takes no part, or at least its place is secondary, in paroxysmal hemoglobinuria. It would seem that alkalization definitely prevents the production of a hemoglobinemia somewhere in the lysin fixation-hemolysis mechanism. Its action is somewhere in the peripheral blood or blood vessels. W. Jacobi (cited by Krogh,[14a] page 178) demonstrated that alkaline solutions placed topically on the web of a frog enhanced the vasoconstrictor action of epinephrine. Although he ascribed this phenomenon to a better absorption of the epinephrine through the frog's epidermis, such could not be the entire case, for with the alkalinity the weak epinephrine solutions constricted vessels on which they ordinarily would have had no effect even when placed directly on them. Could it be that the increased alkalinity in the case of our patient initiated vasoconstriction synergistically with the epinephrine of the blood so that either the endothelial cells could not produce the hemolysin or that the constricted capillaries of the glomeruli would not allow the hemoglobin molecule to pass through with the filtrate? Inasmuch as the absence of hemoglobinuria after the use of sodium bicarbonate, as demonstrated in *B* of table 1, would indicate that increased alkalinity had subdued the influence or the manufacture of the lysin, it is probable that, should an increase in the action of epinephrine be the responsible factor, the efficaciousness of the sodium bicarbonate lies in its synergistic action with epinephrine on the vasoconstrictor apparatus so that the lysin is not

TABLE 2.—*Results of Donath-Landsteiner Tests*

Serum of Patient, Cc.	Cells of Patient, Cc.	Hemolysis	Serum of Patient, Cc.	Cells of Patient, Cc.	Hemolysis
	A			*D*	
0.10	0.50	Trace	0.10	0.50	0
0.20	0.50	1+	0.20	0.50	0
0.30	0.50	1+	0.30	0.50	0
0.50	0.50	2+	0.50	0.50	0
	B			*E*	
0.10	0.50	0	0.10	0.50	Trace
0.20	0.50	0	0.20	0.50	1+
0.30	0.50	Trace	0.30	0.50	1+
0.50	0.50	1+	0.50	0.50	2+
	C			*F*	
0.10	0.50	Trace	0.10	0.50	0
0.20	0.50	1+	0.20	0.50	0
0.30	0.50	1+	0.30	0.50	0
0.50	0.50	2+	0.50	0.50	0

A shows the results of the Donath-Landsteiner test, increasing amounts of the patient's serum being used with a standard amount of a 5 per cent suspension of the patient's washed erythrocytes during the experiment of *A* in table 1. *B* and *C* are the tests for experiments *B* and *C* in table 1. *D* was done soon after sodium bicarbonate in large doses was given for the first time when the urine reached an alkalinity of over p_H 7.5. It was the only time prior to completion of antisyphilitic therapy that the Donath-Landsteiner test was entirely negative. *E* shows the test after half the oxophenarsine hydrochloride and bismuth subsalicylate treatments had been received and *F* the result at the end of the entire course of therapy.

15. Risse, O., cited by Webster and others.[16]

16. Webster, M. D.; Engel, F. L.; Laug, E. P., and Amberson, W. R.: Influence of p_H upon the Elimination of Hemoglobin by a Perfused Frog's Kidney, J. Cell. & Comp. Physiol. **5**:399 (Dec. 20) 1934.

17. Walbum, L. E.: Importance of Hydrogen Ion Concentration in Haemolysis by Lysins of Anaerobic Bacteria, J. Path. & Bact. **46**:85 (Jan.) 1938.

18. De Navasquez, S.: Excretion of Haemoglobin, with Special Reference to "Transfusion" Kidney, J. Path. & Bact. **51**:413 (Nov.) 1940.

produced or that the lysin fixation-hemolysis mechanism is interrupted in the peripheral vessels. In *A* and *C* of table 1, it is clearly demonstrated that erythrocytes, leukocytes and albumin appear in the urine before the hemoglobinuria develops. This would seem to indicate strongly that some lesion is created in the glomerulotubular apparatus, probably by the cold lysin, allowing the excretion of hemoglobin rather than that the hemoglobin caused the renal lesion.

As has been stated, it was Ham's experience [3a] with paroxysmal nocturnal hemoglobinuria that the improvement effected by alkalization of the urine was temporary, there being a recurrence of the hemoglobinuria in spite of continued use of sodium bicarbonate. He also discovered that there was a sharp rise in the hemoglobinuria in his 2 patients soon after the discontinuance of the drug. It is remarkable that the effect of alkalinity should be of some efficaciousness in either case, and it is especially remarkable that the two types of hemoglobinuria should be similarly affected. In Ham's work with the Marchiafavia-Micheli type, the lysin is inherent with the erythrocytes, while with the case of the "cold" hemolysin the lysin is the property of the serum—yet the two types are affected alike by the use of sodium bicarbonate. Although it would seem that alkalinity was the important factor in the changes brought about by the sodium bicarbonate, this statement cannot be made definitely, as the responsible agent may have been the sodium ion. However, Baker and Dodds [13a] showed that hemoglobinuria was easily produced in their experiments when the urine was acidified by the use of sodium sulfate, thus placing the onus on the basic radical. Although this patient had been taking 12 Gm. of sodium bicarbonate daily for thirty days, so that the urine was held at a p_H of 7.5, hemolytic phenomena occurred on the usual refrigeration, and the Donath-Landsteiner reaction was consistently positive (table 1 *C*) in spite of the administration of an additional 20 Gm. forty-eight hours prior to the test. The patient continued his high altitude flying at low temperatures in the interim but had been provided with electrically heated clothing. He had noted no dark-colored urine during this regimen. As soon as it was demonstrated that the use of sodium bicarbonate was of no lasting value and as serotherapy, once thought to be of help,[19] had been proved useless,[20] antisyphilitic therapy was begun with a course of forty 0.06 Gm. doses of oxophenarsine hydrochloride given intravenously and sixteen 0.20 Gm. intramuscular injections of bismuth subsalicylate. As it was felt that any improvement in the patient's hemoglobinuria might be used as a quantitative test in the judgment of the efficacy of this rather short course of therapy, a Donath-Landsteiner test was performed when half the treatment had been finished and at the end of the entire course. *E* and *F* of table 2 show the result of the test at each of these stages of therapy. It is seen that at the halfway point there was no improvement in the test for the lysin, whereas apparent cure as indicated by the negative Donath-Landsteiner test was effected at the end of the treatment. Probably this can be used as another criterion for the efficacy of the short, intensive course of syphilotherapy currently used in the army. No clinical signs of syphilis appeared at any time in this patient, and at the end of his course of therapy the reaction to the Kahn test of the blood was negative and the Wassermann reaction anticomplementary. The results of examination of the spinal fluid, done when he was first seen, were negative.

19. Widal and Rostaine: Sérothérapie préventive de l'attaque d'hémoglobinurie paroxystique, Compt. rend. Soc. de biol. **58**:397, 1905.

20. Montagnami, M.: Crise hémoclasique et hémoglobinurie paroxystique, Presse méd. **29**:1017 (Dec. 24) 1921; cited by Mackenzie.[7]

SUMMARY

A case of paroxysmal hemoglobinuria in an aerial gunner with probable syphilis who was subject to attacks when exposed to the low temperatures of high altitudes is presented. The temperatures to which he was subjected in flying were duplicated in experiments in which the patient was placed in a storage refrigerator, and hemoglobinuria with its attendant signs of blood destruction was observed. Some transient benefit was noted with alkalization of the urine with sodium bicarbonate. The currently prescribed course of forty 0.06 Gm. doses of oxophenarsine hydrochloride administrated intravenously and sixteen 0.20 Gm. doses of bismuth subsalicylate given intramuscularly cured this disease, as evidenced by the development of a negative reaction to a Donath-Landsteiner test. In cases of unexplained anemia, especially but not necessarily in a patient known to be exposed to low temperatures, the phenomenon of paroxysmal hemoglobinuria due to the "cold" hemolysin should be suspected. The search for syphilis by history and by physical examination and serologically (the Wassermann reaction being used in the cases in which the Kahn reaction was negative) is important both as an aid to diagnosis and as an indication for treatment. Some theoretic considerations of the phenomenon of paroxysmal hemoglobinuria are presented.

CONGENITAL IDIOPATHIC METHEMOGLOBINEMIA

FAVORABLE RESPONSE TO ASCORBIC ACID THERAPY

R. F. SIEVERS, M.D., PH.D.
Passed Assistant Surgeon United States Public Health Service
AND
J. B. RYON, M.D.
Senior Surgeon United States Public Health Service
BETHESDA, MD.

The purpose of this report is to describe the observations in a case of congenital idiopathic methemoglobinemia and the favorable response of the patient to ascorbic acid therapy. Methemoglobinemia of a congenital and spontaneous origin is an unusual condition, infrequently reported in the literature. The occurrence of cyanosis because of a high blood level of methemoglobin in two or more members of the same family is referred to as familial idiopathic methemoglobinemia. Among the cases of this type appearing in the literature, the cyanosis has usually been present since the patient's birth but in a few instances has not been observed until later in childhood. This should not be confused with methemoglobinemia of enterogenic origin, such as is described by Stokvis,[1] Talma,[2] van den Bergh,[3] Gibson and Douglas,[4] Lichtenbelt[5] and Wynter,[6] in which the elevated methemoglobin concentration is associated with autointoxication produced by chronic persistent diarrhea or obstipation and which usually disappears when these conditions are brought under control.

Pertinent data on idiopathic methemoglobinemia as reported in the literature are presented in table 1. The first report of congenital cyanosis of long duration in which congenital heart disease and pulmonary disorder were ruled out was that of François,[7] in Belgium, in 1844. It was not until 1912, however, that Slosse and Wybauw,[8] also of Belgium, described idiopathic cyanosis in which the presence of methemoglobin was confirmed by spectroscopic examination. Although their patient had a systolic pulmonic murmur, the cyanosis was thought to be associated with the methemoglobin found in the blood, which disappeared in vitro when a reducing agent was added. This patient, unlike those whose disease is of enterogenic origin, did not have any intestinal trouble; nor did changes in the dietary regimen or purges affect the cyanosis.

The case reports compiled in table 1, with 1 exception, concern patients who were free of gastrointestinal complaints—that is, free from persistent diarrhea or constipation. The 1 exception is the case described by Miller,[9] in which the young girl had obstinate constipation and excessive indicanuria. The daily use of enemas of potassium permanganate solution lessened the degree of cyanosis but failed to efface it completely. The report is included here because the cyanosis dated from birth. Another case in which the mechanism for methemoglobin formation may be different than the others included here is that of Schwartz and Rector.[10] The blood from their patient contained 57 per cent methemoglobin, but no etiologic factors could be ascertained. The baby was given an intravenous injection of a 1 per cent solution of methylthionine chloride. The cyanosis van-

From the Industrial Hygiene Research Laboratory, National Institute of Health.

1. Stokvis, B. J.: Zur Casuistik der autotoxischen enterogenen Cyanosen, Internat. Beitr. z. inn. Med. **1**: 597-610, 1902.

2. Talma, S.: Intraglobulaire methaemoglobinaemie bij den mensch, Nederl. tijdschr. v. geneesk. **38**:721-732, 1902.

3. van den Bergh, A. A. H.: Enterogene Cyanose, Deutsches Arch. f. klin. Med. **83**:86-106, 1905.

4. Gibson, G. A., and Douglas, C. C.: Microbic Cyanosis, Lancet **2**:72-74 (July 14) 1906.

5. Lichtenbelt, J. W. T.: Intraglobulaire Methaemoglobinaemie, Geneesk. gids **1**:545-552, 1923-1924.

6. Wynter, W. E.: Methaemoglobinaemia of Twelve Years Standing, Proc. Roy. Soc. Med. (Clin. Sect.) **1**: 48, 1907-1908.

7. François: Cas de cyanose congénitale sans cause apparente, Bull. Acad. roy. de méd. Belgique **4**:698-700, 1844-1845.

8. Slosse, A., and Wybauw, R.: Un cas de méthémoglobinémie idiopathique, Ann. et. bull. Soc. roy. d. sc. méd. et. nat. de Bruxelles **70**:206-214, 1912.

9. Miller, R.: A Case of Enterogenous Methaemoglobinemia (? Congenital) in a Child, Arch. Dis. Childhood **5**:73-75 (Feb.) 1930.

10. Schwartz, A. S., and Rector, E. J.: Methemoglobinemia of Unknown Origin in a Two Week Old Infant, Am. J. Dis. Child. **60**:652-659 (Sept.) 1940.

ished within thirty minutes and had not returned at the last period of observation, eight months later. The failure of cyanosis to reappear after a single dose of methylthionine chloride, which is lost fairly rapidly by excretion, places this case in a group separate from those just described.

The case reported by Dieckmann [11] is unusually interesting. When first observed, the patient was pregnant for the third time, having had one stillborn child and one who died three months after birth. It was stated that during her pregnancies the cyanosis was more intense. When she went into labor for her third term, she had only 7 Gm. of functioning hemoglobin per hundred cubic centimeters of blood. A transfusion of 600 cc. of whole blood was given, and immediately afterward her skin turned an almost normal color. A normal-appearing baby was delivered without further complications. The cyanotic color did not return during the remaining fourteen days of hospitalization. However, a month after her discharge from the hospital, the cyanosis had reappeared, and a large amount of methemoglobin was found in her blood.

An analysis of these reports reveal (table 1) that 18 cases of idiopathic methemoglobinemia have been recorded to date in the literature. In 8 of these cases the disorder was thought to be of congenital origin only; 6 had both a congenital and a familial history; 2 had a familial history, and in 2 the disease could not be defined as congenital or familial. Only persons of the white race have been described with this disorder. The median age was 29 years, with the entire age range represented. Seven were female, and 11 were males. Nearly all of the cases have been reported since 1930. The entire group may be classified as idiopathic to distinguish these cases from those of methemoglobinemia of enterogenic origin and of other known causes, such as exposure to aniline sulfate or sulfanilamide therapy.

In the majority of the cases just listed, the cyanosis had been attributed to a cardiac defect, usually congenital. Frequently a cardiac murmur was present, and, in some instances, enlargement of the heart was noted. Dyspnea on exertion and tachycardia were frequent findings. There were also complaints of nervousness, irritability, persistent headaches and shortness of breath. Often the skin was described as having a bluish gray color and no underlying reddish tint. The bluish discoloration is par-

TABLE 1.—*Tabulation of Pertinent Data from Reported Cases of Idiopathic Methemoglobinemia*

Nationality	Age, Yr.	Sex	Duration of Cyanosis, Years	Familial History of Cyanosis	Method of Determining Methemoglobin	Amount of Methemoglobin: Gm. per 100 Cc.	Amount of Methemoglobin: Per Cent of Total Hb	Date of Report	Authors
Belgian	20-22	M	20-22	None	.....	—	—	1844	François [7]
Belgian	37	F	4	None	Spectroscopic	—	—	1912	Slosse and Wybauw [8]
Turkish	26	M	7	None	Gasometric and spectroscopic	—	26	1930	Litarczek and others: Sang **4**: 188-202, 1930
British	9	F	9	None	Spectroscopic	—	?	1930	Miller [9]
American	27	F	27	None	Gasometric and spectroscopic	?	?	1932	Dieckmann [11]
German	32	M	32	2 brothers had cyanosis since birth	Gasometric and spectroscopic	—	40	1932	Hitzenberger, K.: Wien. Arch. f. inn. Med. **23**: 85-96, 1932
Dutch	33	F	33	None	Spectroscopic	—	—	1933	van Thienen, G. J.: Nederl. tijdschr. v. geneesk. **77**: 1086-1092 (March 11) 1933
Dutch	16	M	16	None	?	?	?	1933	van Lier, H. W.: Thesis, Utrecht, 1933; cited by van Thienen
German	68	M	68	None	Spectroscopic	—	17	1935	Leiner, G., and Minibeck, H.: Wien. klin. Wchnschr. **48**: 1547 (Dec. 13) 1935
Dutch	37	M	37	Brother and sister had cyanosis	Spectroscopic	—	—	1937	van Heukelom, S.: Geneesk. tijdschr. v. Nederl.—Indië **77**: 3064 (Nov. 30) 1937
Canadian	37	M	25-26	Sister (next case)	Gasometric and spectroscopic	1.4	11.5	1938	Bensley, Rhea and Mills: Quart. J. Med. **7**: 325-330 (April) 1938
Canadian	43	F	32	Brother (above case)	Gasometric and spectroscopic	0.7	5.0	1938	Same as case just above
French	36	M	36	Brother (next case) also 1 brother and 2 sisters	Spectroscopic	—	45	1939	Lian, Frumusan and Sassier [12]
French	28	M	28	See above case	Spectroscopic	—	35-40	1939	Same as case just above
American	2 wk.	F	2 wk.	None	Gasometric and spectroscopic	7.4	57	1940	Schwartz and Rector [10]
British	29	M	29	Brother (next case)	Gasometric and spectroscopic	—	ca. 40	1943	Deeny, Murdock and Rogan [13]
British	19	M	19	Brother (above case)	Gasometric and spectroscopic	—	ca. 40	1943	Same as case just above
British	?	F	Since birth	None	Spectroscopic	6.2	35	1944	King, Gilchrest and White [14]

11. Dieckmann, W. J.: Methemoglobinemia, Arch. Int. Med. **50**:574-578 (Oct.) 1932.

ticularly intense about the lips, ears, nose, cheeks, finger nails and mucous membranes of the mouth. Clubbing of the fingers, which is often associated with certain cardiorespiratory disorders, was not reported. In the cases in which the methemoglobin content was measured the concentration varied from approximately 10 to 57 per cent of the total hemoglobin, the median being between 30 and 40 per cent.

Little has been offered in the literature in regard to the mechanism involved in idiopathic methemoglobinemia. A very important observation was made by Dieckmann [11] when he noted that the methemoglobinemia of his patient remained at a constant level. Ordinarily, methemoglobin will gradually disappear if the blood is allowed to stand in a test tube for twenty-four hours at room temperature. Since the blood cells of his patient were found to have normal glycolytic activity, Dieckmann reasoned that "the constancy of methemoglobinemia, then, is probably due to the continued presence or production of a substance capable of oxidizing hemoglobin." Lian, Frumusan and Sassier [12] discovered that the methemoglobin concentration of the blood of their 2 patients had not changed after the blood had stood in a test tube for twenty-four hours and also that the addition of the patients' plasma to normal red cells did not produce any methemoglobin. These workers suggested that the methemoglobinemia was caused by an abnormal resistance to reduction. They also observed the therapeutic benefits of ascorbic acid administration and demonstrated the demethemoglobinizing property of ascorbic acid with in vitro experiments. One of their patients was given a daily intravenous dose of 100 mg. of ascorbic acid for three weeks, which resulted in a noticeable diminution of cyanosis and methemoglobin concentration, a disappearance of dyspnea and headache and an increased sense of well-being. The ascorbic acid therapy was then discontinued for ten days, during which time the cyanosis again became conspicuous and the methemoglobin concentration rose to 35 per cent.

Unaware of the work of Lian and his group, Deeny, Murdock and Rogan [13] tried oral administration of ascorbic acid. Their first patient, receiving 150 mg. of ascorbic acid twice daily, acquired a normal coloring of the skin in twelve days of treatment. With the same dosage of ascorbic acid continuing to be given, a month later the oxygen-combining capacity of his blood was found to be 22 volumes per cent in comparison to 13.2 volumes (approximately 35 per cent methemoglobin) before treatment. On the sixty-third day of treatment, the patient's blood contained 11 per cent methemoglobin and 1.55 mg. per hundred cubic centimeters of ascorbic acid. The second patient treated by these authors received 200 mg. of ascorbic acid daily for two weeks and then 400 mg. daily thereafter. During the first month of treatment, the methemoglobin concentration of the blood fell from 43 per cent to 6 per cent, at which level it remained. An initial ascorbic acid level of 0.19 mg. per hundred cubic centimeters became stabilized after three weeks of treatment at 1.73 mg. The patient's cyanosis disappeared and strenuous exercise was less exhausting.

A fourth person with idiopathic methemoglobinemia, successfully treated with ascorbic acid, is presented in the report of King, Gilchrest and White.[14] Before treatment, their patient had a blood methemoglobin concentration of approximately 35 per cent and a plasma ascorbic acid level of 0.33 mg. per hundred cubic centimeters. After he had received 300 mg. ascorbic acid per day for one month, the methemoglobin had dropped to 9 per cent, and the plasma ascorbic acid had risen to 1.37 mg. per hundred cubic centimeters. During the second month the daily dose of ascorbic acid was doubled. This did not materially alter the methemoglobin level and raised the ascorbic acid content of the blood only slightly. Approximately one month after the ascorbic acid therapy was discontinued, the methemoglobin concentration rose again to 31.6 per cent, and the plasma level of ascorbic acid returned to the pretreatment level. Gibson [15] was of the opinion that the use of ascorbic acid in the treatment of methemoglobinemia appeared to be limited to those cases in which the normal enzyme systems responsible for reducing methemoglobin were absent or suppressed.

REPORT OF A CASE

During a recent investigation in an ordnance plant, a young woman (B. A.) with pronounced cyanosis, who was working in a nontoxic area, was brought to our attention. She had a dull, gray-purple appearance which was even more conspicuous at short distances (100 ft. [33 meters]) than at close view. B. A. was 19

12. Lian, C.; Frumusan, P., and Sassier: Méthémoglobinémie congénitale et familiale: Action favorable de l'acide ascorbique, Bull. et mém. Soc. méd. d. hôp. de Paris **55**:1194-1203 (July 17) 1939.

13. Deeny, J.; Murdock, E. T., and Rogan, J. J.: Familial Idiopathic Methaemoglobinaemia with a Note on the Treatment of Two Cases with Ascorbic Acid, Brit. M. J. **1**:721-723 (June 12) 1943.

14. King, E. J.; Gilchrest, M., and White, J. C.: A Case of Methemoglobinaemia, Biochem. J. **38**:viii, 1944.

15. Gibson, Q. H.: The Reduction of Methaemoglobin by Ascorbic Acid, Biochem. J. **37**:615-618, 1943.

years old and said she had been cyanotic all of her life. None of her living relatives had cyanosis or other related symptoms. Her father, who had died of a heart attack, was said to have been nervous and had suffered from headaches also. Her color, as well as accompanying symptoms of headaches, extreme fatigue and dyspnea with exercise, had been ascribed to a congenital cardiac lesion by a number of physicians. On physical examination, no evidence of a defective heart could be found; nor could the cyanosis be ascribed to chemical or medicinal exposure or to gastrointestinal disorder. Two samples of dark, chocolate-colored blood were taken about two weeks apart and each found to contain approximately 50 per cent methemoglobin. These levels did not change when the samples remained in a test tube overnight. As congenital methemoglobinemia was suspected, the patient was hospitalized for twenty-four days for investigative purposes six months later.

B. A. had always been in fairly good health except for almost continuous severe headaches, dizziness, dyspnea and palpitation on exertion and frequent sore throats. The headaches occurred in the frontal and occipital areas, were throbbing in character and varied in intensity. The patient stated that they usually developed fifteen to twenty minutes after she arose in the morning and continued with increasing intensity throughout the day. She did not suffer from ocular fatigue or blurred vision. She usually took 4 to 6 acetylsalicylic acid tablets a day but would sometimes total as many as 12. Her headaches were severer when her skin was bluer, and both the headaches and the cyanosis were thought to increase before menstruation. Although the dizziness seemed to be worse after menstruation, she generally felt her best at that time. Sixteen months before observation she had given birth to a normal and healthy-appearing female infant. She believed she was less cyanotic and more energetic during the latter part of her pregnancy than she had ever been.

The patient complained that after moderate exercise her heart beat rapidly and heavily and it was difficult for her to breathe. She said that she could walk only one or two blocks if she moved rapidly but a mile (1.6 kilometers) or more if walking slowly and that she was not able to run at all. Her hands and feet tended to be cold and dry, and her feet swelled occasionally. When she attempted to carry packages or the baby, her arms and hands often became cramped. At the time of observation, she stated she had lost strength during the last three or four years. She admitted that she was an irritable person and became upset easily, frequently quarreling with her family and worrying about finances.

Questioning revealed that the patient's dietary regimen was poor, particularly in protein and vitamins. She usually had a sandwich and a Coca Cola for lunch, about six Coca Colas during the day and a regular evening meal. She took no vitamin capsules and rarely ate raw fruits or vegetables; her appetite had always been good.

B. A. was a short, well developed, slightly obese woman of the stated age. Although there was general cyanosis of her skin, it was particularly noticeable over the nail beds, palms, lower half of both legs, lips and oral mucous membranes. Ophthalmoscopic examination demonstrated extremely dark-colored retinal vessels. There was no clubbing of the fingers.

She had a resting regular pulse rate of 80 per minute, which became decidedly accelerated with mild exercise. The heart sounds were of good quality except for a definite roughness of the pulmonic first sound and an accentuation of the pulmonic second. The average of several readings of blood pressure taken on both arms was 98 systolic and 70 diastolic (millimeters of mercury). The surface area measurement of the heart shadow on a posteroanterior roentgenogram of the chest taken at 72 inches (183 cm.) was of normal size as based on the patient's height and weight. The lungs appeared normal on physical and roentgenographic examination. The neurologic examination showed nothing abnormal except hyperactive tendon reflexes.

Electrocardiograms were taken before and after exercise according to the Master exercise test,[16] but the patient was unable to complete the test because of extreme dyspnea. The tracing taken after exercise showed a slight depression of the ST segment in one of the leads; otherwise the tracing appeared normal.

Hemoglobin and methemoglobin were measured by the spectrophotometric method of Horecker and Brackett.[17] On the day of the patient's admission to the hospital, the methemoglobin content was 3.9 Gm. per hundred cubic centimeters of blood, or about 25.2 per cent of the total hemoglobin. This value was considerably lower than it had been six months earlier. These differences may have been associated with the patient's menstrual cycle, as the first day in the hospital was the fifth and last day of her menstrual period. The results of the laboratory procedures that were performed repeatedly are presented in table 2. Other laboratory observations included the prothrombin time, red cell size and distribution as measured by the Price-Jones technic, plasma fibrinogen content, glycolytic activity of whole blood and twenty-four hour urinary excretion of coproporphyrin, all of which were within normal limits.

Before vitamin therapy was initiated, ascorbic acid was added to the patient's blood in vitro and was found to reduce the methemoglobin. Then the patient was given 100 mg. of ascorbic acid orally, four times daily. During the first seven days of treatment the methemoglobin level gradually dropped from 4.4 Gm. to 2.1 Gm. per hundred cubic centimeters, the methemoglobin falling from 25 to 11 per cent. The ascorbic acid level of the blood rose from 0.5 to 1.5 mg. per hundred cubic centimeters and the urinary level of ascorbic acid from 8.7 to 187.0 mg. per twenty-four hour period, as analyzed by the method of Farmer and Abt.[18] The patient seemed more alert and energetic and stated that her headaches were less intense. The cyanosis was diminished but still conspicuous.

Until this time, use of methylthionine chloride had been avoided because of its two contradictory actions on hemoglobin; that is, low concentrations are known to convert methemoglobin to hemoglobin in vivo, but high concentrations produce methemoglobin by converting the ferrous ion to the ferric. These opposing actions have been explained by the reversible oxidation-

16. Master, A. M.; Friedman, R., and Dack, S.: The Electrocardiogram After Standard Exercise as a Functional Test of the Heart, Am. Heart J. **24**: 777-793 (Dec.) 1942.

17. Horecker, B. L., and Brackett, F. S.: A Rapid Spectrophotometric Method for the Determination of Methemoglobin and Carbonylhemoglobin in Blood, J. Biol. Chem. **152**:699-677 (March) 1944.

18. Farmer, C. J., and Abt, A. F.: Determination of Reduced Ascorbic Acid in Small Amounts of Blood, Proc. Soc. Exper. Biol. & Med. **34**:146-150 (March) 1936.

reduction system of methylthionine chloride and its leukoform.[19] On the eighth day of ascorbic acid therapy, 7 cc. of 1 per cent solution of methylthionine chloride was given intravenously. Within thirty minutes, the remainder of the patient's cyanosis disappeared. A sample of blood taken two hours later contained 0.2 Gm. methemoglobin per hundred cubic centimeters. The patient claimed that her headache had vanished and that she had a sense of well-being. For the first time in her life her skin had a pink color. Her hands and feet were warmer, and she was conscious of her palms becoming damp. She was able to walk rapidly without tiring or becoming breathless. The Master exercise test was repeated, and this time it was completed with ease. The depression of the ST segment following exercise which was found in the electrocardiograms taken before treatment was absent. A comparison of T waves in leads I and II taken at rest, before and after ascorbic acid therapy revealed a slight increase in the height of these waves after treatment. The elevation in the height of the T waves in this case may be associated with the increase in oxyhemoglobin concentration in the blood. The effect of anoxemia, due to decreased atmospheric oxygen tension, on T waves is well known.[20] In addition, similar alterations have been observed by Yonemoto[21] in electrocardiograms taken on rabbits after the blood had been treated with nitrite to produce 68 to 84 per cent methemoglobin.

The methemoglobin started to rise again (table 2) a few hours after methylthionine chloride had been given. The day after this treatment her blood contained 0.65 Gm. per hundred cubic centimeters, or 3.7 per cent methemoglobin. At the time of her discharge from the hospital two days later the methemoglobin content of her blood was 6 per cent and the red blood cell count 5,320,000 per cubic millimeter; the reticulocyte count had dropped to 0.1 per cent, and the total cell volume was 43.5 per cent. The patient's color was normal, and she was entirely symptom free. She was instructed to continue taking 400 mg. of ascorbic acid daily and was also given a prescription for methylthionine chloride (0.3 Gm. by mouth once daily) should she become noticeably cyanotic again. The patient's course has been followed for one year by correspondence with her and with the physician in the plant where she had been employed. The methemoglobin content of her blood as measured on several occasions was approximately 0.7 Gm. per hundred cubic centimeters, and there have been no periods of cyanosis. She has faithfully maintained the high ascorbic acid intake and at no time has resorted to use of methylthionine chloride.

Laboratory Studies on Patient's Blood.—In an effort to determine the cause of idiopathic methemoglobinemia, a series of simple in vitro experiments were performed on the patient's blood before and after ascorbic acid therapy. The results of these procedures may be observed in the chart.

TABLE 2.—*Hematologic Data Before and After Ascorbic Acid Therapy*

Date	Methemoglobin, Gm. per 100 Cc.	Hemoglobin Oxygen, Gm. per 100 Cc.	Total Hemoglobin, Gm. per 100 Cc.	Methemoglobin, per Cent of Total Hemoglobin	Total Cell Volume, per Cent	Ascorbic Acid: Plasma, Mg. per 100 Cc.	Ascorbic Acid: Urine, Mg. per 24 Hr.	Blood Count: Red Blood Cells, Millions per Cu. Mm.	Blood Count: White Blood Cells, Thousands per Cu. Mm.	Blood Count: Reticulocytes, per Cent
11/ 4/43	3.90	11.60	15.50	25.2	....	....	....	5.46	7.0	1.8
11/ 6/43	3.80	11.10	14.90	25.5	44.7	....	....	5.58	6.8	
11/ 9/43	4.20	10.95	15.15	27.7	....	....	4.8	....	...	1.4
11/12/43	4.30	13.90	18.20	23.6	....	0.40	7.3			
11/15/43	4.40	13.20	17.60	25.0	45.5	0.51	8.7	5.39	8.5	0.1
11/16/43	Started ascorbic acid treatment—400 mg. per day									
11/17/43	3.10	15.00	18.10	17.1	....	....	....	....	...	0.8
11/18/43	2.50	15.20	17.70	14.1	....	1.18	67.8	5.39	7.4	1.0
11/19/43	2.40	16.45	18.85	12.7	....	....	174.2			
11/20/43	2.35	15.80	18.15	13.0	....	1.58	158.4			
11/21/43	....	.....	.....	....	....	....	187.0			
11/22/43	2.10	16.00	18.10	11.6						
(9:30 a. m.)*	2.30	14.60	16.90	13.6	....	1.58	327.7			
11/23/43 (11:00 a. m.)	0.20	17.42	17.62	1.1						
11/23/43 (3:00 p. m.)	0.40	—	—	—						
11/24/43	0.65	16.85	17.50	3.7						
11/25/43	0.60	14.70	15.30	3.9						
11/26/43	1.08	15.70	16.78	6.2	43.5	....	.....	5.32	...	0.1

* Intravenous injection of 7 cc. of 1 per cent solution of methylthionine chloride.

Samples of the patient's blood taken on different days were allowed to remain in a test tube at room temperature, and determinations of methemoglobin were made at intervals of three to twelve hours. Repeated analyses over periods of twenty-four to seventy-two hours failed to demonstrate any significant change in the methemoglobin concentration. This finding is in contrast to changes that occurred in normal blood to which sodium nitrite had been added to produce methemoglobin. Normal blood, with intact red cells, methemoglobinized to approximately 8 Gm. per hundred cubic centimeters by the addition of 0.31 mg. sodium nitrite per cubic centimeter lost most of its methemoglobin in a twenty-four hour period (section *A* in the chart). The rate of conversion of methemoglobin to hemoglobin at room temperature in the normal nitrited blood samples which were analyzed during this study averaged 0.3 Gm. per hundred cubic centimeters per hour. The reduction of methemoglobin in hemolyzed normal blood also occurs at a rapid rate. Within seventy-two hours there was no change in the methemoglobin content of the patient's hemolyzed blood (section *B* in the chart).

The next experiment concerned the influence of plasma of another source on the patient's corpuscular

19. Hauschild, F.: Die Wirkung des Katalysins (Thionin) bei der Methämoglobinvergiftung, Arch. f. exper. Path. u. Pharmakol. **184**:458-467, 1937.

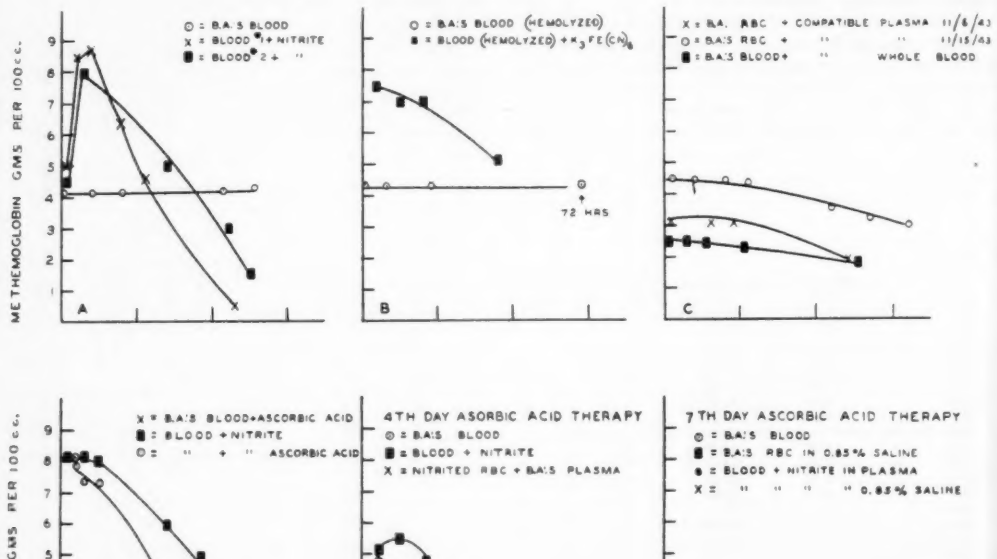
20. White, M. S.: Effect of Anoxia in High Altitude Flights on the Electrocardiogram, J. Aviation Med. **11**:166-180 (Dec.) 1940.

21. Yonemoto, G.: Electrocardiogram During Methemoglobinemia Produced by Injection of Sodium Nitrite, J. Orient. Med. **35**:117-126, 1941.

methemoglobin content. The formed elements of the patient's blood were separated by centrifugation, and the plasma was drawn off and replaced with an equal amount of compatible plasma. There was practically no change in the methemoglobin levels during the first eight to ten hours in two separate tests. A reduction did take place in both tests during the tenth to twenty-fifth hours, but the rate of conversion did not approach that which occurs in normal nitrited blood. The compatible plasma contained 1.69 mg. per hundred cubic centimeters of ascorbic acid in comparison with the patient's plasma, which contained 0.4 and 0.5 mg. During the twenty-four hour period, the erythrocytes in contact with compatible plasma lost approximately 1.3 Gm. of methemoglobin per hundred cubic centimeters of blood (section *C* in the chart).

Compatible whole blood was then added to the patient's whole blood, which diluted the amount of methemoglobin by one half at the onset of the experiment. The methemoglobin concentration of the blood mixture was reduced from 2.5 to 1.9 Gm. per hundred cubic centimeters in twenty-three hours, which is much less than the reduction to be expected with nitrited blood.

The effectiveness of ascorbic acid as a reducing agent was studied on both normal blood treated with sodium nitrite and on the patient's blood. To the nitrited blood containing 8.5 Gm. methemoglobin per hundred cubic


METHEMOGLOBIN GMS PER 100 C.C.
○ = B.A.'S BLOOD
X = BLOOD + NITRITE
■ = BLOOD + 2 + "
A
○ = B.A.'S BLOOD (HEMOLYZED)
■ = BLOOD (HEMOLYZED) + K3 FE(CN)6
72 HRS
B
X = B.A. RBC + COMPATIBLE PLASMA 11/6/43
○ = B.A.'S RBC + " " 11/15/43
■ = B.A.'S BLOOD + " WHOLE BLOOD
C
METHEMOGLOBIN GMS PER 100 C.C.
X = B.A.'S BLOOD + ASCORBIC ACID
■ = BLOOD + NITRITE
○ = " + " ASCORBIC ACID
D
TIME IN HOURS
4TH DAY ASORBIC ACID THERAPY
○ = B.A.'S BLOOD
■ = BLOOD + NITRITE
X = NITRITED RBC + B.A.'S PLASMA
E
TIME IN HOURS
7TH DAY ASCORBIC ACID THERAPY
○ = B.A.'S BLOOD
■ = B.A.'S RBC IN 0.85% SALINE
● = BLOOD + NITRITE IN PLASMA
X = " " " " 0.85% SALINE
F
TIME IN HOURS

In vitro studies at room temperature of reduction of methemoglobin in oxalated blood from patient (B. A.) and from a normal subject treated with methemoglobin formers. *A*, rate of reduction of methemoglobin in B. A.'s blood before ascorbic acid therapy and in two samples of normal blood treated with sodium nitrite (0.31 mg. per cubic centimeter of blood). *B*, the effect of hemolysis on the rate of methemoglobin reduction in B. A.'s blood and in normal blood plus 1.48 mg. potassium ferrocyanide ($K_3 Fe[CN]_6$) per cubic centimeter of blood. (The hemolyzing solution consisted of a borate buffer, p_H 9.4, containing 0.3 Gm. of saponin per hundred cubic centimeters.) Two tenths cubic centimeter of blood was diluted to 2 cc. with the hemolyzing solution. *C*, the effect of compatible plasma and compatible whole blood on the reduction of methemoglobin in B. A.'s blood. In two samples taken nine days apart the plasma was separated from the erythrocytes and replaced with an equal amount of compatible plasma. To a third sample an equal amount of compatible whole blood was added. *D*, the effect of addition of ascorbic acid on methemoglobin reduction in B. A.'s blood in vitro and in normal blood treated with sodium nitrite (0.31 mg. per cubic centimeter). Four-tenths mg. ascorbic acid per cubic centimeter was added to the sample of B. A.'s blood and to one half of the nitrited sample. The other half of the nitrited sample served as a control. *E*, rate of methemoglobin reduction in B. A.'s blood on the fourth day of ascorbic acid therapy and in nitrited erythrocytes plus B. A.'s plasma. Two hours after addition of sodium nitrite (0.15 mg. per cubic centimeter) the plasma was separated from one half of the sample and replaced with an equal volume of B. A.'s plasma. *F*, seventh day of ascorbic acid therapy. Rate of methemoglobin conversion of B. A.'s blood and a portion of her blood in which the plasma has been replaced with an equal volume of 0.85 per cent sodium chloride solution; also, comparison of normal nitrited blood (0.08 mg. sodium nitrite per cubic centimeter) in which the plasma from one half of the sample was separated and replaced with an equal volume of 0.85 per cent sodium chloride solution.

centimeters and to the patient's blood were added 0.4 mg. of ascorbic acid per cubic centimeter of blood. This amount was stoichiometric for the methemoglobin content of the nitrited blood and a two-fold excess for the patient's blood; it also represented fifty times the amount of ascorbic acid ordinarily present in the plasma. The methemoglobin in the patient's blood sample was rapidly converted to hemoglobin, and the rate of methemoglobin reduction in the nitrited sample was accelerated (section *D* in the chart).

The next series of test tube experiments were conducted on the patient's blood on the fourth day of ascorbic acid therapy. At this time analysis immediately after withdrawal of venous blood showed that the methemoglobin content was 2.5 Gm. per hundred cubic centimeters, a drop of 1.9 Gm. in four days of treatment. When this blood sample was allowed to stand in a test tube, the methemoglobin concentration continued to fall to 0.9 Gm. per hundred cubic centimeters in twenty-four hours. During this same twenty-four hour period in vivo methemoglobin concentration dropped only 0.1 Gm. per hundred cubic centimeters.

The relative effectiveness of B. A.'s plasma in reducing the methemoglobin of nitrited blood was compared, after four days of vitamin therapy, with that of normal plasma. A sample of nitrited blood was divided into equal portions and the plasma of one portion replaced with the patient's plasma. Unfortunately, the oxidation reaction of the nitrite was incomplete when the plasma was separated for replacement, and methemoglobin continued to increase in the control sample. Taking this factor into account, the rate of reduction of methemoglobin continued at very nearly the same rate in both samples. The ascorbic acid levels for the two plasma samples were 1.2 mg per hundred cubic centimeters for the patient and 1.7 mg. for the control (section *E* in the chart).

The role of plasma in methemoglobin reduction was next studied by comparing the effectiveness of the patient's plasma on the seventh day of therapy with that of isotonic solution of sodium chloride. Half of a blood sample from the patient was centrifuged and the plasma replaced with 0.85 per cent sodium chloride solution. The other half was used as the control. Measurements were then made of the reduction rate of the methemoglobin in each preparation for a period of twenty-four hours. The methemoglobin disappeared at the same rate in both samples. A second test was made with normal nitrited blood which was similarly handled. The conversion of methemoglobin to the reduced form again progressed at nearly the same rate when the cells were suspended in plasma or in sodium chloride solution (section *F* of the chart). In addition, the plasma was removed from a sample of the patient's blood, and twenty-eight hours later the cells were made up to the former volume with sodium chloride solution. A determination made immediately revealed only a small quantity of methemoglobin.

COMMENT

These experiments indicate that before ascorbic acid therapy the patient's blood had little or no power to reduce methemoglobin to hemoglobin in vitro. This is in contrast to the action of normal nitrited blood, in which methemoglobin was reduced at the average rate of 0.3 Gm. per hundred cubic centimeters per hour. The separation of the patient's erythrocytes and the addition of compatible plasma containing a high normal amount of ascorbic acid (1.7 mg. per hundred cubic centimeters) caused a reduction in methemoglobin of only 1.3 Gm. over a period of twenty-four hours. The delayed onset of reduction on the addition of compatible plasma suggests that a transfer of a reducing substance into the erythrocytes may have taken place. After four days of ascorbic acid therapy, the methemoglobin content of the patient's blood in vitro was reduced 1.6 Gm. per hundred cubic centimeters within twenty-four hours. The patient's plasma at this time contained 1.2 mg. of ascorbic acid per hundred cubic centimeters. Since, stoichiometrically, 10 mg. of ascorbic acid per hundred cubic centimeters, or eight times the concentration present in the plasma, would be required to reduce 1.6 Gm. of methemoglobin per hundred cubic centimeters, it becomes apparent that either another reducing system has been activated or the ascorbic acid itself is acting catalytically. The equal rates of disappearance of methemoglobin from the erythrocytes of normal nitrited blood when suspended in their own plasma or when suspended in a 0.85 per cent solution of sodium chloride indicate that the constituents of the plasma are not primarily responsible for the conversion of methemoglobin to hemoglobin. This is emphasized by the repetition of this action when the patient's erythrocytes were suspended in an isotonic solution of sodium chloride on the seventh day of ascorbic acid therapy, at which time the patient's plasma contained 1.58 mg. of ascorbic acid per hundred cubic centimeters. The patient's red cells probably contained a comparable concentration of ascorbic acid.

These experiments suggest that the conversion of methemoglobin to hemoglobin is dependent on the reducing systems present within the red cell. Cox and Wendel[22] were of the opinion that the reduction of methemoglobin was mainly, if not entirely, a function of the enzyme system contained within the erythrocyte. In the present case a partial failure of one of the reducing systems within the erythrocyte was suspected. By elevating the ascorbic acid content of the blood to unusually high levels, the defective reducing system was partially replaced or reactivated, and the methemoglobin content of the blood was reduced, both in vivo and in vitro. The reducing mechanism was still imperfect, because a high ascorbic acid intake (over 400 mg. per day) did not reduce

22. Cox, W. W., and Wendel, W. B.: The Normal Rate of Reduction of Methemoglobin in Dogs, J. Biol. Chem. **143**:331-340 (April) 1942.

the methemoglobin level below 0.5 to 0.6 Gm. per hundred cubic centimeters. Although the patient's methemoglobin concentration was temporarily reduced to 0 after the intravenous injection of methylene blue, it then leveled off at about 1.0 Gm. per hundred cubic centimeters, which is approximately eight to ten times the average concentration found in a normal person.[23] This failure to reduce the methemoglobin concentration to normal levels may be noted in 4 other patients with idiopathic methemoglobinemia successfully treated with ascorbic acid. In no case did the methemoglobin level drop to normal, but each remained at a concentration between 6 to 11 per cent of the total pigment.

Judging from the experience of the small number of patients treated, it appears that a high concentration of ascorbic acid in the blood will control methemoglobinemia of idiopathic origin. The plasma levels of ascorbic acid which were recorded in 3 cases, including this one, were, before treatment: 0.19,[13] 0.31[14] and 0.40 to 0.51, mg. per hundred cubic centimeters (this case). After ascorbic acid therapy was initiated and before a reduction in the methemoglobin content had become noticeable, these levels had risen to 0.46, 1.17 and 1.24, mg. per hundred cubic centimeters, respectively. In the case reported by King, Gilchrest and White, the methemoglobin in the circulating blood began to increase when the plasma level of ascorbic acid had dropped to 0.61 mg. per hundred cubic centimeters after the discontinuation of ascorbic acid therapy.

In normal blood, ascorbic acid probably does not play an essential role in the maintenance of hemoglobin in a functionally active state. For example, there are no known reports of methemoglobinemia among persons with as low plasma contents of ascorbic acid as occur in scurvy. However, from the evidence presented in this case and from several reports in the literature, it would appear that there is a concentration of ascorbic acid in the blood that is critical in the treatment of methemoglobinemia of idiopathic origin.

The methemoglobin concentration is probably regulated by the various oxidation-reduction systems present in the red blood corpuscle. Warburg and Christian[24] have shown with their experiments on the various enzyme systems that methemoglobin is reduced by the system hexosemonophosphate - dehydrogenase - triphosphopyridinenucleotide. The oxidation-reduction potential of the methemoglobin-hemoglobin system of the horse, when the concentration of oxidant and reductant are equal, is + 0.139 volt at *p*H, 7.0 at 30 C.[25] Normally, human blood contains approximately 1 per cent of total hemoglobin as methemoglobin. By computation[26] the potential for 1 per cent methemoglobin and 99 per cent hemoglobin is about 0 volt. Our patient's blood before treatment contained from 25 to 50 per cent methemoglobin as measured on widely separated occasions. The calculated potential for 25 per cent methemoglobin and 75 per cent reduced hemoglobin is + 0.11 volt. On the basis of this calculation it necessarily follows that the normal reducing systems were inactive.

The following data reported in the literature may be relevant to this concept. The various normal values recorded for the potential of whole blood in vitro range from + 0.110 to + 0.200 volt, depending on the procedure used for measuring the potential.[27] The presence of intact erythrocytes has but slight influence on the potential of plasma or serum. However, when the blood is hemolyzed there is a definite drop in potential, which indicates that the potential within the normal red cell is much lower than that in the plasma.[28] On the other hand, hemoglobin is converted to methemoglobin when added to plasma. In the circulating blood as much as 40 per cent of injected hemoglobin may be converted to methemoglobin over a period of twenty-four to forty-eight hours.

23. Paul, W. D., and Kemp, C. R.: Methemoglobin: A Normal Constituent of Blood, Proc. Soc. Exper. Biol. & Med. **56**:54-55 (May) 1944. Sievers, R. F.; Lawton, A. H.; Skoog, F.; Neal, P. A.; von Oettingen, W. F.; Stump, R. L., and Monaco, A. R.: A Medical Study of the Effect of TNT on Workers in a Bomb and Shell Loading Plant, Public Health Bulletin 291, United States Treasury Department, Public Health Service, 1945.

24. Warburg, O., and Christian, W.: Ueber Aktivierung der Robisonschen Hexose-Mono-Phosphosäure in roten Blutzellen und die Gewinnung aktivierender Fermentlösungen, Biochem. Ztschr. **242**:206-227 (Nov. 17) 1931.

25. Taylor, J. F., and Hastings, A. B.: Oxidation-Reduction Potentials of the Methemoglobin-Hemoglobin System, J. Biol. Chem. **131**:649-622 (Dec.) 1939.

26. $E_h = E_o \frac{0.060}{\eta} \times \log \frac{\text{oxidant}}{\text{reductant}}$ (Bodansky, M.: Introduction to Physiological Chemistry, New York, John Wiley & Sons, Inc. 1938.)

27. Meier, L.: Die Bestimmung des Redoxpotentials im Blut (in vitro) und die Beeinflussung durch Zufuhren sogenannter Redoxsubstanzen, Biochem. Ztschr. **303**:32-39 (Nov.) 1939. Oivine, I. A.: Potential oxydo-réducteur (E_h) du sang, Bull. biol. med. exper., U. R. S. S. **3**:542-544, 1937.

28. Oivine, I. A.: Controversial Problems of the Oxidation-Reduction Potential of the Blood, Bull. biol. med. exper., U. R. S. S. **7**:344-347, 1939.

On the basis of this evidence, the following hypothesis is offered as a suggestion for the mechanism involved in idiopathic methemoglobinemia:

Methemoglobin is constantly being produced in the circulating blood. In the case of idiopathic methemoglobinemia the reducing systems within the erythrocyte which normally keep methemoglobin at low concentrations are inactive or at reduced activity. In the absence of these reducing systems the potential of the erythrocyte will tend to increase as the diffusible oxidation-reduction systems of the plasma enter the cell, resulting in the production of methemoglobin. The extent of methemoglobin formation will be a function of the degree to which the normal reducing systems are lacking in the erythrocyte and of the rate at which the plasma constituents will diffuse into the cell. In the treatment of idiopathic methemoglobinemia with large doses of ascorbic acid, the high concentrations of ascorbic acid within the erythrocyte serve as a substitute for the missing reducing system.

SUMMARY

Medical and laboratory tests and observations were made on a 19 year old woman with methemoglobinemia dating from birth. There was no evidence of gastrointestinal disturbance, and no other member of her family had cyanosis.

The observations reported by others that the oral administration of ascorbic acid diminishes methemoglobinemia and cyanosis were confirmed. It was found that relatively high plasma concentrations of ascorbic acid were required to be effective and that the methemoglobin was not reduced to the levels found in normal circulating blood. Complete and temporary reduction of methemoglobin after an intravenous injection of methylene blue with return of methemoglobin concentration to approximately 6 per cent while the patient had a high ascorbic acid intake was demonstrated.

In vitro experiments were conducted prior to and during treatment in an attempt to determine the mechanism of idiopathic methemoglobinemia. From results obtained in these experiments a hypothesis was formulated to explain this mechanism.

STUDY OF WATER AND HEAT LOSS FROM THE RESPIRATORY TRACT OF MAN

METHODS: I. A GRAVIMETRIC METHOD FOR THE MEASUREMENT OF THE RATE OF WATER LOSS; II. A QUANTITATIVE METHOD FOR THE MEASUREMENT OF THE RATE OF HEAT LOSS

GEORGE E. BURCH, M.D.
NEW ORLEANS

In any study of water loss from the human body it is necessary to measure that which leaves the respiratory system. Methods have been described, but they are either inaccurate or cumbersome. It is beyond the scope of this paper to review the methods used in the past to measure the water loss through the pulmonary system; many of these are referred to in several excellent discussions on insensible perspiration.[1] Galeotti and his associates [2] and Weyrich [3] used bottles of sulfuric acid or calcium chloride to trap the expired water and measure gravimetrically the water collected. These are not simple procedures, and their accuracy is not well established. The observers did not control the conditions of the air inspired. The paper by Galeotti and Signorelli [2a] included a review of some of the early methods used to measure expired water. Loewy and Gerhartz [4] measured the temperature of the air expired but not the water loss from the lungs. The method of Benedict and Benedict,[5] using chemical absorption, is rather cumbersome and relatively inaccurate. Christie and Loomis [6] employed an ingenious and accurate method for measuring the water vapor pressure of expired air. The method cannot be employed to determine accurately the quantity of water loss from the lungs. In an excellent and more recent paper Seeley [7] described a method using anhydrous magnesium perchlorate in U tubes to collect and weigh the expired water. Not only is this method cumbersome, but its accuracy is not fully established. It was intended and was used by Seeley with success for the measurement of relatively small samples (1 to 2 liters) of expired air. In another study [8] I like Benedict and Benedict,[5] Newburgh and Johnston [1c] and others, employed a large sensitive balance to measure water loss from the lungs and skin. This method is relatively inaccurate and not applicable to certain types of studies.

In order to study water and heat loss from the lungs it became necessary to develop an accurate, dependable and practical method for measuring water loss. The method was developed further to measure heat loss simultaneously with water loss in a manner so that ambulatory and hospital patients as well as normal subjects could be studied in large numbers and at frequent intervals.

To facilitate presentation, the paper is divided into two parts, describing (1) the method for measuring water loss gravimetrically and (2) the

This is the thirteenth paper from the Laboratory of Tropical Physiology.

Aided by a grant from the Rockefeller Foundation and the Helis Institute for Medical Research.

From the Department of Medicine, Tulane University School of Medicine and Charity Hospital of Louisiana.

1. (*a*) Soderstrom, G. F., and Du Bois, E. F.: Clinical Calorimetry: XXV. The Water Elimination Through Skin and Respiratory Passages in Health and Disease, Arch. Int. Med. **19**:931 (May) 1917. (*b*) Benedict, F. G., and Root, H. F.: Insensible Perspiration: Its Relation to Human Physiology and Pathology, ibid. **38**:1 (July) 1926. (*c*) Newburgh, L. H., and Johnston, M. W.: The Insensible Loss of Water, Physiol. Rev. **22**:1, 1942. (*d*) Hill, L.: The Science of Ventilation and Open Air Treatment, London, His Majesty's Stationery Office, 1919.

2. (*a*) Galeotti, G., and Signorelli, E.: Ueber die Wasserbilanz während der Ruhe und bei der Anstrengung im Hochgebirge, Biochem. Ztschr. **41**:268, 1912. (*b*) Galeotti, G.: Ueber die Ausscheidung des Wassers bei der Atmung, ibid. **46**:173, 1912.

3. Weyrich, W.: Beobachtungen über die unmerkliche Wasserausscheidung der Lungen und ihr Verhältniss zur Hautperspiration, Dorpat, E. J. Karow, 1865.

4. Loewy, A., and Gerhartz, H.: Ueber die Temperatur der Expirationsluft und der Lungenluft, Arch. f. d. ges. Physiol. **155**:231, 1914.

5. Benedict, F. G., and Benedict, C. G.: Perspiratio insensibilis: Ihr Wesen und ihre Ursachen, Biochem. Ztschr. **186**:278, 1927.

6. Christie, R. V., and Loomis, A. L.: The Pressure of Aqueous Vapour in the Alveolar Air, J. Physiol. **77**:35, 1932.

7. Seeley, L. E.: Study of Changes in the Temperature and Water Vapor Content of Respired Air in the Nasal Cavity, Tr. Am. Soc. Heat & Vent. Engin. **46**:259, 1940.

8. Burch, G. E.: A Method for Measuring Small Amounts of Weight Loss in Man, Am. J. M. Sc., to be published.

method for measuring heat loss from the respiratory tract.

I. A GRAVIMETRIC METHOD FOR THE MEASUREMENT OF EXPIRED WATER

The apparatus and method developed to measure quantitatively the expired water are illustrated diagrammatically by figure 1. The apparatus consists essentially of three parts: (1) two meters for measuring the volume of air circulated through the lungs and collecting coils, (2) aluminum collecting coils and (3) a system of rubber tubing with necessary heater, flutter valves and mouthpiece to direct the flow of air.

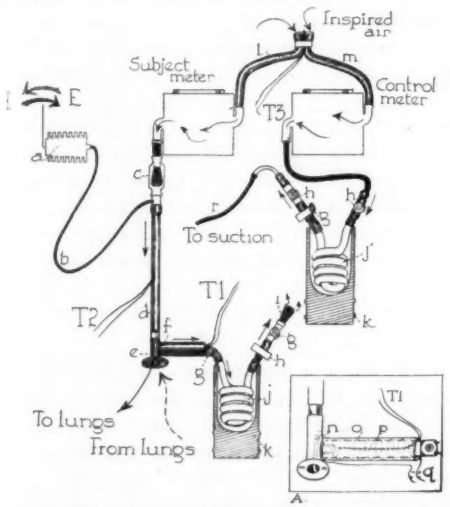


Fig. 1.—A schematic representation of the apparatus employed to measure the rate of water loss from the respiratory tract. The mouthpiece (*e*) is placed in the subject's mouth, and his nose is clipped closed. On breathing he inspires room air through tubing (1). The air passes through the "subject meter," where its volume is recorded. It then passes through the flutter valve and housing (*c*) and through rubber tube (*d*) into his lungs. On expiration the air passes out through the side tubing in the mouthpiece through the heated rubber efferent tube (insert *A*) into aluminum coil (*j*) and out through the trap (*h*), stopcock (*g*) and flutter valve (*i*) into the atmosphere.

Copper—constantan thermocouples (T_1, T_2 and T_3) are used to record the temperature of the inspired and expired air. The brass bellows (*a*) with pivoted lever is connected to the tubing (*d*) by means of rubber tubing (*b*) and indicates mechanically the phases of respiration.

Room air is drawn by means of a suction system through rubber tubing (*m*) and through the control meter, where its volume is recorded. It is then drawn through aluminum coil (*j*), through the trap and then out into the suction system by means of tubing (*r*).

Insert *A* shows the nature of the construction of the electrically heated efferent tubing carrying the expired air to the aluminum coil. A wall (*n*) is constructed in each mouthpiece to prevent the inspired and expired air from mixing outside the subject's mouth.

1. The meters are ordinary commercial test gas meters which measure the volume of air flow under low pressure to an accuracy of about 30 cc. All air that circulates through the lungs and coils passes through the meters so that the volume and rate of circulation of air may be determined.

2. Aluminum coils are constructed and employed essentially as described for the coils used for measuring water loss from the skin.[9] The coils are made to weigh less than 200 Gm., so that they can be weighed on an ordinary analytic balance. The internal diameter of the aluminum coils is about 16 mm. and the walls about 0.7 mm. thick. The length of tubing in each coil is about 160 cm. There is no kinking of the tubing, and the diameter of the coils is small enough for them to be placed on an ordinary analytic balance. A finished coil is shown in figure 2. Each open end of the coil is connected by rubber tubing to an aluminum stopcock, to make it possible to isolate the lumen of the collecting coil from the

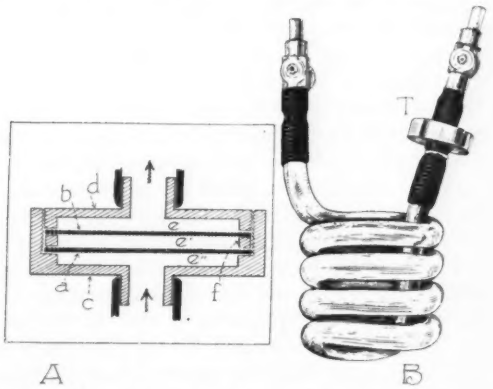


Fig. 2.—A schematic representation of the aluminum collecting coils. Insert *A* shows the details of construction of the trap (*T*) used to prevent the escape of frozen water on expiration. Parts *a* and *b* are fine meshed brass filters; *e*, *e'* and *e''* are air spaces between the filters which allow free circulation of the expired air.

atmosphere. By carefully varying the amount of rubber tubing and aluminum tubing slightly, all coils are made to weigh about the same (within a few milligrams) to facilitate weighings.

A metal filter (insert *A*, fig. 2) is placed just proximal to the stopcock guarding the exit from the coils, to prevent any of the condensed frozen expired water from being blown out of the coils.

9. Neumann, C.; Cohn, A. E., and Burch, G. E.: A Quantitative Method for the Measurement of the Rate of Water Loss from Small Areas, with Results for Finger Tip, Toe Tip, and Posterio-Superior Portion of the Pinna of Normal Resting Subjects, Am. J. Physiol. **132**:748, 1941.

This filter consists of a short broad thin-walled cylindric aluminum housing and two layers of 100 mesh per square inch brass screen. The two layers of screen filter are kept apart from each other by means of a metal ring (*f*, insert *A*, fig. 2). The diameter of the housing is 4.0 cm. and the length 1.0 cm. The large diameter reduces the resistance to the flow of air through the housing, and the double layer of filter insures complete trapping of the snow.

3. The rubber tubing, flutter valves and mouthpiece are connected as shown in figure 1. A nichrome heater (*o*, insert *A*, fig. 1) surrounds the efferent tube (*p*) leading from the mouthpiece to the subject collecting coil [10] to maintain the walls of this efferent tubing at a temperature of 92 F. (33.4 C.) to prevent condensation of expired water within its walls. A rheostat is connected in series with the heater to vary the heater temperature whenever the environmental temperature is varied.

In use, the previously dried (by fanning with an electric fan and by wiping the external surface and drawing air through the interior) and weighed coils are placed in a thermos jar and iced with chips of solid carbon dioxide (fig. 1). This procedure cools the coils to about —70 C. The coils are then connected to the meters, the patient and the air suction lines as indicated by figure 1. The room in which the studies are conducted is air conditioned. The subject, who has rested a given period, inserts the mouthpiece (such as is used for ordinary clinical measurements of metabolic rate) in his mouth and adjusts his chair and the apparatus so that he is comfortable and well relaxed. He is instructed to breathe through his nose while all stopcocks are opened and air lines through the subject coil made patent. The nose is then closed with a clip, and timing with a stopwatch is started simultaneously. The subject then inhales air from the room through the subject meter (fig. 1) into his lungs, out through the warm efferent tube (*p*) and through the subject coil (*j*), where the expired water condenses, the dry air escaping into the atmosphere. This procedure is continued for a period of five minutes.

Simultaneously with the collection of the expired water from the subject, an equal volume of room air is drawn through the room meter and through the room coil (*j*), the dry room air escaping into the vacuum system providing the suction. The intake openings for the subject meter and the room meter are placed adjacent to each other in order to insure an entrance of similar room air into both systems, that of the patient and that of the control, or room air, system. After a given period (accurately recorded) of study, the stopcocks guarding the collecting coils are closed. The coils are dried by fanning and wiping and are weighed again. The gain in weight represents the quantity of water collected over the known period of time and after known volumes of air have irrigated the respiratory tract. From the known quantities, the subject's weight and height, the grams of water collected and the time, the rate of water loss is calculated, expressed in grams per square meter of body surface per ten minutes.

The number of grams of water collected in the subject coil represents (1) the water in the room air inspired and (2) the water lost from the respiratory tract. The water collected in the room coil represents the water contained in the room air inspired. The difference between the number of grams of water collected in the subject coil and that collected in the room coil represents the quantity of water lost from the respiratory tract.

CALIBRATION AND TESTING OF APPARATUS AND METHOD

1. *Accuracy of the Method.*—To determine the accuracy of the method a Kjeldahl connection bulb containing water was placed in the air line of the subject coil and meter in the relative position occupied by the subject. Air was drawn through the subject meter, through the connection bulb containing a known amount of water and then through the subject coil. Simultaneously an equal volume of room air was drawn through the room meter and coil. By weighing the coils and connection bulb before and after this procedure, it was possible to determine the accuracy of the method for collecting expired water. It was possible to vary the quantity of water lost by the connection bulb by heating the bulb.

The results are shown in the table. The mean error is —0.27 per cent and the extremes + 1.25 and — 1.27 per cent. The results were not influenced by the quantity of water collected within the limits of amounts involved when subjects were studied. More rapid rates of water loss were not studied. Variations in the rate of air flow within the limits encountered when studying subjects did not influence the accuracy of the method.

2. *Completeness of Collection of Water by a Single Collecting Coil.*—In order to determine the completeness with which the water is trapped by the subject coil, two coils were connected in series and iced with solid carbon dioxide. Room

10. The term "subject coil" is employed to indicate the aluminum collecting coil used to trap the expired water and "room coil" to indicate the collecting coil used to measure the water content of an equal volume of room air.

air was drawn through the coils, or a subject expired through the coils. It was found that the first coil through which the moisture-laden room air or expired air entered trapped the moisture almost completely. For example, when the quantity of water trapped by the first coil was about 400 mg., the maximum amount of water collected by the second collecting coil in the series did not exceed 4 mg. When the first coil collected about 1,500 mg. of water, the second coil collected a maximum of 17 mg. These figures would seem to indicate that when quantities of water collected

The Accuracy of the Proposed Method for Measuring the Rate of Water Loss From the Respiratory Tract

Test No.	Known Water Loss, Gm.	Water Collected, Gm.	Error, Gm.	Error, per Cent
1	0.7943	0.7899	−0.0044	−0.55
2	1.0578	1.0553	−0.0025	−0.24
3	0.7657	0.7646	−0.0011	−0.14
4	0.7653	0.7602	−0.0051	−0.67
5	0.7064	0.7152	+0.0088	+1.25
6	0.8163	0.8112	−0.0051	−0.62
7	0.8596	0.8656	+0.0060	+0.70
8	1.0781	1.0730	−0.0051	−0.47
9	1.0821	1.0743	−0.0078	−0.72
10	0.8337	0.8231	−0.0106	−1.27
Mean.....	0.87593	0.87324	−0.0027	−0.27
Maximum...	1.0821	1.0743	+0.0088	+1.25
Minimum...	0.7064	0.7152	−0.0106	−1.27

are near 1 Gm. or less the error due to incomplete removal of the water from the expired air is less than 1 per cent. It will be seen in the papers to follow that in the usual experiment the water collected in any one collecting coil rarely exceeds 1 Gm.

3. *The Control of Water in Room Air.*—The accuracy of the method depends on the accuracy with which the water in the room air is measured. For example, room air is drawn through the two meters and collecting coils simultaneously and through the orifices of two rubber tubes placed adjacently (fig. 1). It is possible that the water contents of air entering each differ. To test this possibility, room air of equal volume was drawn through a separate meter and coil simultaneously.

In a typical experiment, one coil collected 0.4686 Gm. of moisture from the room air, and the other collected 0.4685 Gm. from an equal volume of room air. These amounts indicate that the method employed for controlling the moisture in the room air is satisfactory.

4. *Errors Involved in Drying and Weighing the Collecting Coils.*—The process of drying the outside of the coils by wiping and fanning and of weighing, icing them, drying them again by wiping and fanning and then weighing them again must entail certain errors. As a test, several coils were put through the process outlined in the preceding sentence. The results showed an error not exceeding 2 mg. and usually amounting to less than 1 mg. This is a surprisingly small error considering the surface area of the coils, the method of drying and the accuracy of the balance (± 0.1 mg.) used for the weights.

5. *Resistance to Air Flow Through the Coils.*—The length of the aluminum tubing and the diameter of the lumen has increased the friction to the flow of air from the respiratory tract on expiration. By placing a T tube in the system of tubing near the mouthpiece and connecting this to a mercury manometer, it was found that the pressure of expiration on quiet respiration averaged + 36 mm. of water (2.6 mm. of mercury), the extremes being 30 and 40. The pressure on inspiration was of essentially the same magnitude but negative. When inspiration or expiration was sudden or jerky, the pressure changes were slightly greater.

6. *Condensation of Moisture in the System Other than in the Collecting Coils.*—The first group of experiments showing the accuracy of the method indicated the lack of any appreciable loss of water by condensation in the system other than in the collecting coils. The heater around the efferent tubing prevented any such loss. As will be seen, a thermocouple placed within the efferent tube made it possible to be sure that sufficient warmth of the walls of the efferent tube existed prior to the start of any study in order to prevent condensation on its walls. Furthermore, segments of glass tubing were placed in various places within the system. The failure of the walls of the glass tube to be clouded by water condensing on their surfaces after prolonged breathing through the system indicated that no large quantities of water were lost by condensation within the tubings.

II. METHOD FOR MEASURING THE RATE OF HEAT LOSS FROM THE RESPIRATORY TRACT

With only relatively slight modifications, the method just described for determinations of water loss may be used to measure the heat loss from the lungs simultaneously with the water loss.

Heat is lost from the lungs by (1) evaporation of water, (2) warming of inspired air (convection) and (3) decomposition of carbonic acid in solution to carbon dioxide gas with expiration. The first is measured by the method just described, therefore making it necessary to measure only two other factors, the kilogram calories lost by (1) warming inspired air and (2) the excretion of carbon dioxide.

The Method for Measuring Heat Loss by Warming of Inspired Air (Convection).—To measure the heat loss by warming of inspired air

thermocouples were placed in the efferent tube (p), in the afferent tube (d) and at the site of entrance of the room air into the subject meter (fig. 1). These made it possible to determine with a potentiometer (accurate to 0.1 degree [C.]) the temperature of the inspired and expired air. The subject meter recorded the volume of inspired air in a known period of time. From the temperature of the expired air and from the number of grams of water removed from the expired air, it was possible to determine the mass of air expired as well as its specific heat. With these factors known it was possible to calculate the rate of heat loss by convection.

The fine wire used for the thermocouples (copper-constantan) made it possible to record the temperature of the expired air during expiration as the expired air passed over the thermocouple junction. The heat from the nichrome heater around the efferent tube did not interfere with the recording accurately of the temperature of the expired air, because of the heater's being outside a thick-walled rubber tubing (rubber is

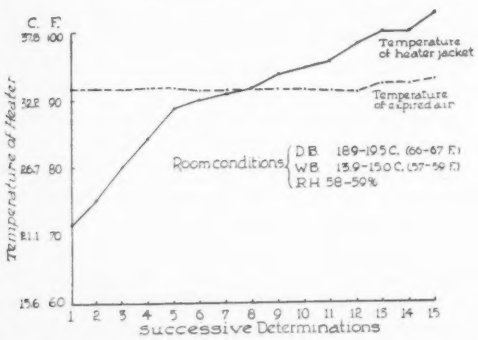


Fig. 3.—The influence of the temperature of the heater jacket and efferent tube (insert *A*, fig. 1) on the temperature of the expired air. The conditions of the room (temperatures by dry bulb and wet bulb thermometers and relative humidity) are shown.

a good insulator of heat), because of the fineness of the thermocouple wire and because of the setting of the temperature of the heater at a level very near (0 to 1 C. variation) the temperature of the expired air. Figure 3 shows that the temperature of the heater did not influence the recording of the expired air unless the temperature of the heater exceeded 36 C. (about 96 F.). For heater temperatures as low as 21 C. (about 70 F.) the recording of the expired air temperature was not altered.

The temperatures are recorded from two to four minutes after the subject begins to respire in the water-collecting apparatus. It does not take nearly as long as two minutes for a state of equilibrium to be reached, but to avoid errors it is preferable to wait at least two minutes.

The Measurement of Heat Loss by Excretion of Carbon Dioxide.—The excretion of carbon dioxide was measured by having the subject breathe in a Benedict-Roth type of clinical machine for measuring the basal metabolic rate immediately after collecting the expired water, measuring inspired and expired air temperatures and so on. This was done without allowing the subject to move from his previous sitting position used for measuring the expired water. Since the subjects were all normal adults on a mixed diet the rate of liberation of carbon dioxide was calculated from the rate of absorption of oxygen as recorded by the B.M.R. machine. This calculation was made by using a respiratory quotient of 0.83, that is, CO_2 (liters) $= 0.83\ O_2$ (liters).

At the same time the recorded metabolic rate (M.R.), not basal metabolic rate (B.M.R.), was determined. Since the subjects rested from thirty to sixty minutes in the observation room before the metabolic rate was recorded, it is fairly safe to assume that the body was in temperature equilibrium with the environment and that, therefore, the total body heat production (determined from the metabolic rate) was equal to the total heat loss. This assumption certainly is true when the subject is in heat equilibrium with his environment. From this measured heat loss it is possible then to express heat loss by all or any of the three previously mentioned components of respiratory heat loss as per cent of total body heat loss.

By measuring the subject's height and weight it is possible to correct for body size. This is done by expressing all heat loss in units of kilogram calories (large or physiologic) per square meter of surface area per ten minutes.

MATHEMATICAL CONSIDERATION OF HEAT LOSS FROM THE RESPIRATORY TRACT

The heat loss from the respiratory tract, as previously mentioned, consists of three components. It does not include heat loss by conduction or radiation, two factors important in heat loss from the surface of the body. The loss of heat [11] from the lungs may be expressed as

$$H = h_E + h_C = h_{CO_2}, \qquad (1)$$

where

H = Total heat loss from the respiratory tract
h_E = Heat loss by evaporation of water from the membranes of the respiratory tract
h_C = Heat loss by warming inspired air (convection); (this may be a positive number if air warmer than body temperature is inspired)

11. Units for heat loss henceforth will be kilogram calories per square meter of surface area of body per ten minutes.

h_{CO_2} = Heat loss by virtue of the decomposition of H_2CO_3 in solution to CO_2 gas, which is expired.

The heat loss by evaporation of water may be expressed as

$$h_E = \frac{0.575\ W}{A} \qquad (2)$$

where

h_E = kilogram calory per square meter of surface area per ten minutes

W = grams of water lost from the respiratory tract per ten minutes

A = surface area of the subject in square meters

The parameter 0.575 is the part of a kilogram calory necessary to vaporize 1 Gm. of water at a temperature of 37.25 C.

The heat lost by convection (heating of inspired air) may be expressed as

$$h_C = \frac{M\ (t^1 - t)\ S}{A} \qquad (3)$$

where

h_C = kilogram calory per square meter of surface area per ten minutes

M = mass of air expired (in grams) in ten minutes

t^1 = temperature of the expired air in degrees Centigrade

t = temperature of the inspired air in degrees Centigrade

S = specific heat of air

A = surface area of the subject's body in square meters.

Equation (3) becomes (for dry air)

$$h_C = \frac{0.2404\ M\ (t^1 - t)}{1000\ A} = \frac{2404\ M\ (t^1 - t)}{10^7\ A} \qquad (4)$$

The heat lost by the excretion of CO_2 may be expressed at standard conditions as

$$h_{CO_2} = \frac{L\ h_V}{22.4\ A} \qquad (5)$$

where

h_{CO_2} = kilogram calory per square meter of surface area per ten minutes

L = liters of CO_2 expired in ten minutes

h_V = kilogram calory of heat absorbed per mol of CO_2 gas liberated when H_2CO_3 in solution is decomposed

A = surface area of the subject's body in square meters

22.4 = volume of a mol of a gas under standard conditions

Equation 5 becomes

$$h_{CO_2} = \frac{4.7\ L}{22.4\ A} \qquad (6)$$

From the thermochemical equations, the decomposition of carbonic acid in an infinite dilution to water and carbon dioxide is associated with the absorption of 4.7 kilogram calories for each mol of carbon dioxide liberated. The blood in the lungs is not truly an infinite dilution of carbonic acid, but for practical purposes it may be considered as such.

From equations (2), (4) and (6), equation (1) becomes

$$H = \frac{0.575\ W}{A} + \frac{2404\ M\ (t^1 - t)}{10^7\ A} + \frac{4.7\ L}{22.4\ A} \qquad (7)$$

COMMENT

The method for measuring expired water is accurate and simple to use. It can be used in the study of subjects when exercising, sitting or lying. Variations in the conditions of the room air do not influence the results, since the water in the room air is collected and measured simultaneously with the expired water. The pressure of 30 to 40 mm. of water against which the subjects must expire is an objection and may be eliminated when the proper type of aluminum tubing becomes available. That, however, is not a serious objection, since the pressure is not high and is fairly constant.

The methods used by others [12] have not been accurately calibrated and in many instances not calibrated at all. The completeness of absorption of the expired air has not been carefully determined. All of them offer a certain amount of resistance to expiration, a factor not quantitively expressed in the published reports.

Sulfuric acid has been one of the agents frequently employed to absorb the expired moisture. It does not lend itself well for such measurements, since a relatively large volume of sulfuric acid must be used to absorb completely the moisture in the expired air. Such large volumes are difficult to weigh accurately during short time studies as well as dangerous to use.

In the use of cold aluminum coils, obstruction of the lumen by frozen water has not occurred in a single instance. Of course, it is necessary to make certain that the interior of the coils is dry and that the studies do not extend over too long a period of time. Five minute studies were conducted in observations made in the Laboratory of Tropical Physiology. As shown in figure 2, a filter was inserted proximal to the stopcock at the exit of the coil in order to prevent any of the snow or frozen water from being blown out of the coil. There has been no plugging of the filter during the studies in this laboratory.

The methods for the measurement of heat loss from the respiratory tract and the partition into the three components, (1) heat loss by evaporation of water, (2) heat loss by warming of inspired air and (3) heat loss by excretion of carbon dioxide, are not complicated. Of the three, the measurement of heat loss by evapo-

12. Hill.[1d] Galeotti and Signorelli.[2a] Galeotti.[2b] Weyrich.[3] Loewy and Gerhartz.[4] Benedict and Benedict.[5]

ration of water is most accurate. In a group of studies, measurement of water loss from the lungs is accurate to about 0.3 per cent without altering the practicability of the measurements. The measurements of heat loss by the other two factors (h_c and h_{CO_2}) may be made more accurate by the use of more sensitive but less practical methods than those outlined. The accuracy of the methods presented is great enough, and the methods are still simple and practical enough for use in the study of many normal and diseased subjects. The results of such studies are presented in detail in another paper.[13]

It is obvious that more satisfactory measurements of heat loss can be made in a room with its temperature and relative humidity controlled, such a room being employed in studies reported from the Laboratory of Tropical Physiology.[13] This is not necessary, however, since all the measurements can be made within an hour, during which time the atmospheric conditions of most observation rooms are constant, and it is then necessary only to measure and describe the conditions of the room during the study.

13. Burch, G. E.: The Rate of Water and Heat Loss from the Respiratory Tract in Normal Subjects in a Subtropical Climate, to be published.

SUMMARY

A simple and practical gravimetric method was devised for the measurement of water loss from the respiratory tract. The mean accuracy of the method is about 0.3 per cent and the extremes ± 1.3 per cent. The use of the method was studied.

A method was devised for the measurement of heat loss from the respiratory tract. The components of the heat loss were measured separately.

Mr. G. Morgavi assisted in these studies.

RATE OF WATER AND HEAT LOSS FROM THE RESPIRATORY TRACT OF NORMAL SUBJECTS IN A SUBTROPICAL CLIMATE

GEORGE E. BURCH, M.D.
NEW ORLEANS

The rate of water loss and heat loss from the lungs of man has not received much attention during the past two decades. Most of the studies were conducted in the latter part of the last century and the early part of this one.[1] In studies of insensible perspiration, water loss from the lungs was measured indirectly by a large sensitive balance, a method originated by Sanctorius.[2] More recently Seeley [3] and Christie and Loomis [4] have studied the temperature of expired air and some aspects of expired water. Except for the early studies, there have been no detailed observations of late devoted to a quantitative measurement of expired water and to the study of factors influencing this loss. There are considerable discrepancies in the early papers. For example, Weyrich [1d] stated that he had definitely shown that expired air is completely saturated with water vapor. Galeotti,[1g] on the other hand, found pulmonary air at body temperature to be only partially saturated at the temperature of expired air. It is known, however, that it is impossible completely to saturate inspired air with water from the surfaces of the respiratory tract, for the water on these surfaces contains colloid and crystalloid materials. The early studies in the literature also failed to consider adequately the conditions of the inspired air, the nature of respiratory conditions of the patient and many other factors concerned with the elimination of water by respiration. Seeley,[3] in an excellent report, considered many of the factors influencing the loss of water by the lungs. His studies should have been extended further, to include more subjects and more quantitative data. Studies in a subtropical climate have been neglected. As was pointed out previously, it is relatively simple to measure the heat loss with an evaluation of its various components while measuring the rate of water loss from the respiratory tract.[5]

This report is concerned with (1) measurements of the rate of water loss from the respiratory tract of normal young adults, (2) study of factors influencing this water loss and (3) measurements of the rate of heat loss from the respiratory tract, with its partition into three main factors. All subjects lived in a subtropical climate and were studied under various conditions of room temperature and relative humidity so as to simulate tropical, desert and cool environments. It was the purpose of this study to measure these physiologic phenomena in a subject sitting quietly, not in a basal state, under known environmental conditions, the attempt being made to duplicate conditions of any man merely sitting restfully in his home.

METHODS AND MATERIAL

The methods used for the measurements of water and heat loss from the respiratory tract have been

From the Department of Medicine, Tulane University School of Medicine and Charity Hospital.

Aided by a grant from the Helis Institute for Medical Research and the Rockefeller Foundation.

1. (*a*) Galeotti, G., and Signorelli, E.: Ueber die Wasserbilanz während der Ruhe und bei der Anstrengung im Hochgebirge, Biochem. Ztschr. **41**:268, 1912. (*b*) Loewy, A., and Gerhartz, H.: Ueber die Temperature der Expirationsluft und der Lungenluft, Arch. f. d. ges. Physiol. **155**:231, 1914. (*c*) Benedict, F. G., and Benedict, C. G.: Perspiratio insensibilis: Ihr Wesen und ihre Ursachen, Biochem. Ztschr. **186**: 278, 1927. (*d*) Weyrich, W.: Beobachtungen über die unmerkliche Wasserausscheidung der Lungen und ihr Verhältniss zur Hautperspiration, Dorpat, E. J. Karow, 1865. (*e*) Rubner, M.: Notiz über die Wasserdampfausscheidung durch die Lunge, Arch. f. Hyg. **33**:151, 1898. (*f*) Galeotti, G.: Wassergehalt und Temperatur der ausgeatmeter Luft, Arch. f. d. ges. Physiol. **160**:27, 1914-1915; (*g*) Ueber die Ausscheidung des Wassers bei der Atmung, Biochem. Ztschr. **46**:173, 1912. (*h*) Loewy, A., and Gerhartz, H.: Ueber die Ausscheidung des Wasser bei der Atmung, Biochem. Ztschr. **47**:343, 1912.

2. Benedict, F. G., and Root, H. F.: Insensible Perspiration: Its Relation to Human Physiology and Pathology, Arch. Int. Med. **38**:1 (July) 1926. Newburgh, L. H., and Johnston, N. W.: The Insensible Loss of Water, Physiol. Rev. **22**:1, 1942.

3. Seeley, L. E.: Study of Changes in the Temperature and Water Vapor Content of Respired Air in the Nasal Cavity, Tr. Am. Soc. Heat. & Vent. Engin. **46**:259, 1940.

4. Christie, R. V., and Loomis, A. L.: The Pressure of Aqueous Vapour in the Alveolar Air, J. Physiol. **77**:35, 1932.

5. Burch, G. E.: A Study of Heat and Water Loss from the Respiratory Tract of Man. Methods: I. A Gravimetric Method for the Measurement of the Rate of Water Loss; II. A Quantitative Method for the Study of the Rate of Heat Loss, this issue, p. 308.

described.[5] All 107 subjects studied varied from 17 to 43 years of age unless otherwise stated. Except for 4 subjects, all were less than 30 years of age and only 3 were less than 20 years (one 17 years and two 19). There were 8 older subjects (fig. 1) for whom only the rate of water loss was measured. The subjects were normal and experienced around laboratories and included both sexes and the white and Negro races. They were eating an average American diet and were free from any mild infections. The subjects were studied from two to four hours postprandially. The observations were made in an air-conditioned room. Unless otherwise stated, the room was comfortable to practically all subjects; that is, the temperature varied from 20 to 21.1 C. (68 to 70 F.) and the relative humidity from 55 to 60 per cent.[6] The subjects who considered the room a little cool were allowed to put on a light jacket to provide comfort. Any variations in the room conditions from those described will be noted.

The studies were conducted in New Orleans during the months of August through December 1944 and January 1945.

All subjects rested, sitting quietly from twenty to forty-five minutes, usually thirty minutes, in comfortable chairs before the observations were begun. They entered into peaceful conversation with the observers and shifted in their chairs as desired. They were not in a basal state. The room was specially constructed to reduce psychic influences from noises, excessive laboratory apparatus and the like. Only one or two observers were present at any one time.

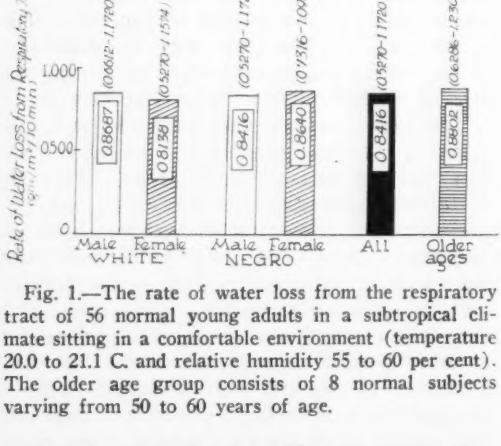


Fig. 1.—The rate of water loss from the respiratory tract of 56 normal young adults in a subtropical climate sitting in a comfortable environment (temperature 20.0 to 21.1 C. and relative humidity 55 to 60 per cent). The older age group consists of 8 normal subjects varying from 50 to 60 years of age.

After the rest period, the subject was connected to the apparatus. During a period of five minutes the water and heat loss were measured. A sling psychrometer was used from one to three times during the five minute period to determine the conditions of the room air. At the completion of the five minute period the subject was allowed to rest for about ten minutes, and then the rate of his total body heat production, elimination of carbon dioxide, volume of tidal air and respiratory rate were recorded with a clinical type of Benedict-Roth B.M.R. machine. For many subjects the measurements were repeated at intervals varying from a few minutes to days or weeks.

For the sake of clarity and continuity of thought the various experiments and results are grouped separately for presentation and comment.

I. THE RATE OF WATER LOSS FROM THE RESPIRATORY TRACT

During two hot summer months (August and September 1944) in New Orleans the mean rate of water loss from the respiratory tract for an entire group of 56 normal young adults varying in age from 17 to 43 years was 0.842 Gm. per square meter of surface area of the body per ten minutes,[7] the extremes being 0.527 and 1.172 Gm. The detailed values for the sexes and for the two races are shown in figure 1. There was

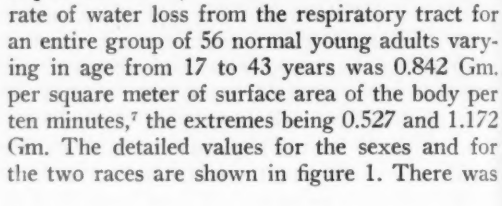


Fig. 2.—A representation of the high correlation of the rate of water loss from the respiratory tract and the rate of irrigation of the respiratory tract with air in 56 normal young adults sitting in a comfortable environment (temperature 20.0 to 21.1 C. and relative humidity 50 to 60 per cent).

no significant difference due to sex or race. The statistical constants for the rate of water loss were:

Mean, 0.878 ± 0.030 Gm. per square meter per ten minutes

Standard deviation, 0.333 ± 0.021 Gm. per square meter per ten minutes

Coefficient of variation, 37.90 ± 2.49 per cent.

The mean rate with which the respiratory tract was irrigated with room air was 38.326 liters per square meter of surface area per ten minutes, the extremes being 28.316 and 53.250 liters. There was no significant difference due to sex or race. The statistical constants for the rate with which

6. Henceforth, a comfortable room will mean one with the air temperature and relative humidity as indicated.

7. The rate of water loss is expressed henceforth in grams per square meter of surface area of the body per ten minutes. All values, when indicated, were expressed according to surface area and ten minute intervals of time.

the respiratory tract was irrigated with air were:

Mean, 38.286 ± 0.933 liters per square meter per ten minutes

Standard deviation, 10.216 ± 0.651 liters per square meter per ten minutes

Coefficient of variation, 26.68 ± 1.82 per cent

The individual values for the rate of water loss and the rate of irrigation of the respiratory tract with room air are shown in figure 2. There is a high positive correlation between these two variables. This is borne out by statistical analysis, the correlation coefficient being + 0.914 ± 0.015.

The Influence of Season.—During the relatively cool month of January these observations were repeated on 27 normal young adults (8 were the same subjects studied during the summer and 19 were different) to study seasonal influences. The conditions of the experiments were the same as for the summer. There were no significant seasonal differences noted. The mean rate of water loss from the respiratory tract was 0.804 Gm. per square meter per ten minutes, with the extremes being 0.508 and 1.205 Gm. The mean rate of irrigation of the respiratory tract with air was 35.577 liters per square meter per ten minutes, the extremes being 26.733 and 49.211 liters. The rate of water loss and irrigation of the respiratory tract with air correlated very highly again, the correlation coefficient being + 0.878 ± 0.030. Thus it can be seen that there was no significant seasonal influence on the rate of water loss from the respiratory tract.

The Influence of Exercise.—The influence of mild exercise on water loss from the respiratory tract was studied in 5 normal young adults. In each subject the rate of water loss was measured as previously described, and then each was made to run in place with moderate rapidity for a period of ninety seconds, and the measurements were repeated. The effects of mild exercise on the rate of water loss, rate of irrigation of the respiratory tract with air and temperature of the expired air are summarized briefly by figure 3. The temperature of the expired air was not changed or was only slightly reduced by exercise, there being an average drop of only 0.3 degree (C.). The rate of water loss from the respiratory tract was definitely increased by exercise, an increase of 36.3 per cent being recorded. The rate of irrigation of the respiratory tract with air likewise was increased, an increase of 45.1 per cent being noted. The increase in the rate of water loss is to be expected, since exercise is known to increase the rate of irrigation of the lungs with air and, as just noted, there is a direct and high correlation of the rate of water loss from the respiratory tract with the rate of irrigation of the respiratory tract with air.

The Influence of Breathing Through the Nose or Mouth on the Rate of Water Loss.—In the method employed the subjects breathed through the mouth. Normally most breathing is through the nose. To learn whether or not breathing through the nose or mouth would influence the results, measurements were made for five subjects, under identical conditions, breathing through the nose and then through the mouth. Special nose pieces were used for the nose. No significant difference was noted in the rate of water loss. For example, for 1 subject the rate of water loss through the nose was 0.6945 Gm. per square meter per ten minutes and through the mouth 0.6903 Gm. per square meter per ten minutes. The temperature of the expired air was essentially the same with both routes of breathing, averaging 33.6 C. for the nasal route and 34.4 C. for the oral route. There was complete overlapping of the variations. This conclusion is not surprising in view of the results of Seeley [3] and Christie and Loomis.[4] These observers showed that the greatest amounts of water and heat are added to inspired air in the alveoli, in which there is more surface area and, therefore, more water and more heat in direct contact with the air. Whether or not the same results would be encountered in long experiments is not known. These studies lasted only five minutes.

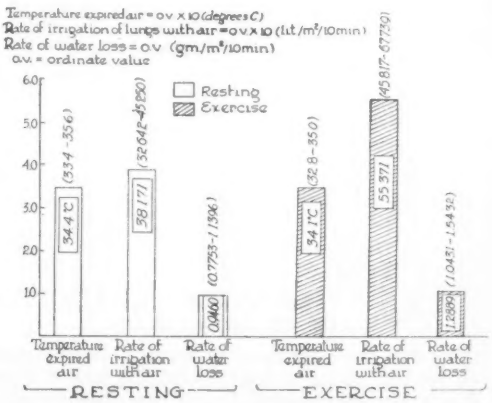


Fig. 3.—The effects of mild exercise on temperature of the expired air, rate of water loss and rate of irrigation of the respiratory tract with air in 5 normal young adults.

then with slow and deep breathing. No attempt was made to quantitate the rates or depths of breathing except to make certain that they were different.

The results are shown in figure 4. The rate of water loss from the respiratory tract essentially doubled with rapid and shallow breathing over that with normal quiet breathing and increased about 1.8 times with slow and deep breathing. There was a more or less comparable increase in the rate of irrigation of the respiratory tract with air. The quantity of air irrigating the respiratory tract and not the nature of the breathing was the principal factor which determined the rate of water loss. However, the data

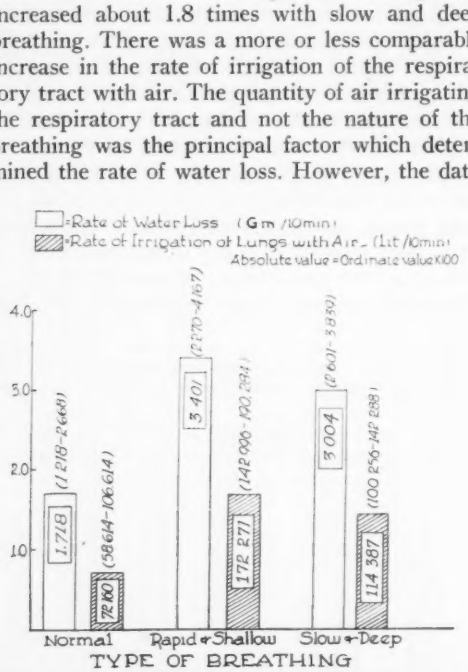


Fig. 4.—The influence of respiratory rate and depth on the rate of water loss. Again, the rate of water loss is determined mainly by the rate of irrigation of the respiratory tract by air.

(fig. 4) show that more water is lost from the lungs per unit volume of air inspired with deep and slow breathing than with rapid and shallow. This greater water loss is probably due to the fact that the air remains in the lungs for a longer time with deep breathing, thus allowing more time for the water on the surface of the respiratory tract to evaporate and to diffuse into the inspired air before it is expired.

The Effect of Room Temperature and Humidity (Cool and Dry, Cool and Foggy, Comfortable, Hot and Dry and Hot and Wet) on Water Loss.—Fourteen normal white adults (23 to 40 years old and all except 1 being female) were studied to learn the influence of variations in the room air on the heat and water loss from the respiratory tract. Some of the measurements were made on different days, since it was not possible to study the influence of all room conditions in immediate succession.

The results, including the absolute values, are summarized in figure 5 for rate of water loss, rate of irrigation of the lungs with air and the temperature and relative humidity of the expired air. The room conditions are indicated by the figure. The temperature of the expired air, the relative humidity of the expired air, the rate of water loss from the respiratory tract and the rate of irrigation of the respiratory tract with air were not significantly different when the subjects were breathing air in a cool dry room, a cool foggy room or a comfortable room (fig. 5). When the room temperature was increased to about 50 C., the rate of water loss did not change significantly, but there was a significant increase in the temperature of the expired air and in the rate of irrigation of the lungs with air. The temperature of the inspired air (room air) was increased over that for the comfortable room conditions by about 30 C., while the temperature of the expired air increased only about 6 degrees (C.). This difference indicated a significant absorption of heat by the respiratory tract, with a cooling of the inspired air by the respiratory tract by about 10 C.

In the hot humid atmosphere there was a decided drop in the rate of water loss from the respiratory tract and a definite drop in the relative humidity of the expired air, while the temperature of the expired air and the rate of irrigation of the respiratory tract increased slightly over that in a hot and dry atmosphere. It is obvious that in a hot and humid atmosphere the rate of water loss does not obey the same laws governing the relation with the rate of irrigation of the respiratory tract with air.

These findings deserve special comment. That the rate of water loss from the respiratory tract is not significantly different when an atmosphere of cool dry, cool foggy or a comfortable air is breathed is to be expected, since the absolute quantities of water vapor per unit volume of air at 15 C. and 60 per cent relative humidity, 15 C. and 94 per cent relative humidity or 20 C. and 52 per cent relative humidity are not very different, the values being 7.9, 12.4 and 12.7 mg. per liter, respectively.[8] When it is remembered that expired air has a temperature of about 33 C. and a relative humidity of about 88 per cent and therefore contains 31.6 mg. per liter, it can be seen that large and essentially equal amounts of water can be and are added to the inspired air under the three room conditions before it is expired.

8. Marvin, C. F.: Psychrometric Tables, United States Department of Agriculture, Weather Bureau, 1915.

When hot (much above body temperature) air is inspired, the problem becomes different. For example, when hot dry air, 50 C. and 18 per cent relative humidity, containing 13.3 mg. per liter, is inspired and merely cooled by the respiratory tract to about 39 C., as has been indicated, the relative humidity of the inspired air suddenly increases to about 38 per cent, still containing 13.3 mg. per liter. This air is capable of taking on more water than cool air, but its dryness and its ability to remove water from the respiratory passage are not as great as might be expected at a first glance, that is, when erroneously considering the hot dry air inspired without the effects of pure. Furthermore, the vapor pressure of air at 39 C. and 80 per cent relative humidity is extremely high, and the water vapor would therefore be forced out of the nose or mouth into the atmosphere with much lower vapor pressures before high values of relative humidity could be reached. In a hot and humid environment, then, the rate of water loss from the respiratory tract would be expected to decrease, while in a cool and foggy air (relative humidity high and absolute amount of water relatively low) the rate of water loss would not be expected to be significantly different from that in a comfortable environment.

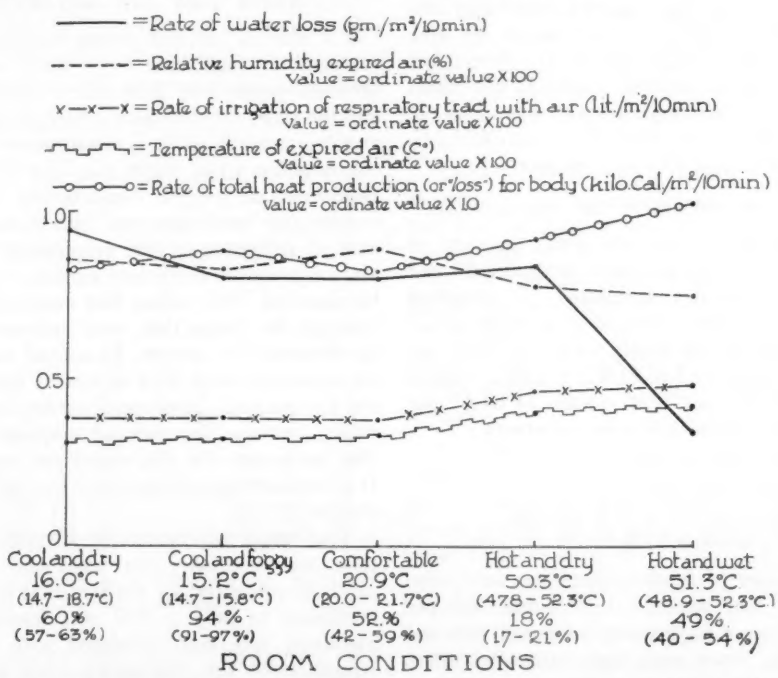


Fig. 5.—A graphic representation of the influence of room temperature and humidity on rate of water loss, temperature of expired air, rate of irrigation of lung with air, relative humidity of the expired air and total heat production.

cooling by the respiratory tract. This effect of cooling on hot humid air (50 C. and 49 per cent relative humidity) is even more pronounced. When such air, containing 37.1 mg. of water per liter, is merely cooled by the respiratory passage to 39 C., its relative humidity is raised to 80 per cent, but it still contains the same 37.1 mg. of water per liter of air, so that little water can be added to this air by the respiratory tract. This phenomenon was verified and is indicated in figure 5. Again, it is impossible to reach a 100 per cent relative humidity in expired air, because the water on the surface of the membranes of the respiratory tract is not chemically

II. TEMPERATURE OF EXPIRED AIR

The temperature of the expired air measured with a thermocouple in 27 sitting normal young adults was found to vary between 31.6 and 34.2 C. when the air inspired had a mean temperature of 20.5 C. (extremes 19.5 and 21.4 C.) and a mean relative humidity of 57.2 per cent (extremes 54 and 61 per cent). The statistical constants found were:

Mean, 33.19 ± 0.21 C.
Standard deviation, 1.58 ± 0.15 C.
Coefficient of variations, 4.76 ± 0.44 per cent

As reported previously,[5] the temperature of the expired air did not vary significantly with

the temperature of the inspired air when it was as low as 20 C. and as high as 36 C. When air temperatures higher than 36 C. were inspired, the temperature of the expired air increased.[5] Weyrich [1d] found the temperature of the expired air to change little even when the temperature of the inspired air was as low as 0 C. The temperature of the expired air reported in the literature varies a great deal in some instances, but most of the reports [9] agreed with the values found in these studies.

When the subjects inspired air from a cool dry atmosphere, the mean temperature of the expired air was 33.1 C., the extremes being 31.7 and 34.2 C. When they inspired cool foggy air, the mean temperature of the expired air was 32.9 C. (extremes 32.2 and 33.6). When hot dry or hot moist air was inspired, the mean temperature of the expired air was 39.6 C. (extremes 38.1 and 40.6 C.) and 42.1 C. (extremes 40.6 and 43.4 C.) respectively.

III. RELATIVE HUMIDITY OF EXPIRED AIR

The temperature and the total quantity of water in the expired air were determined [8] and the relative humidity calculated. A statistical analysis of the data collected in a study of 27 normal young sitting adults breathing room air at 20.5 C. (range 19.5 to 21.4 C.) with a relative humidity of 57.2 per cent (range 54 to 61 per cent) resulted in the following constants:

Mean, 88.15 ± 1.31 per cent
Range, 78 to 96 per cent
Standard deviation, 9.87 ± 0.92 per cent
Coefficient of variation, 11.20 ± 1.06 per cent

Three values of relative humidity were relatively high; they were 95.0, 95.8 and 96.5 per cent. These were high when compared with the other values. Since such high values were not found when the determinations were repeated, they therefore may have been in error.

A mean value of 88 per cent for the relative humidity of the expired air is consistent with the known physical and physiologic facts. It is physically impossible to obtain a 100 per cent relative humidity in expired air, since the water on the surfaces of the respiratory tract is not chemically pure. Claims of finding complete saturation of expired air with water have been made by many observers.[10] These claims are invalid because of obvious errors.

9. Weyrich.[1d] Galeotti.[1g] Seeley.[3]

10. (*a*) Osborne, W. A.: Water in Expired Air, J. Physiol. **47**:12, 1913. (*b*) Luciani, L.: Human Physiology, New York, The Macmillan Company, 1913, vol. 1, p. 397. (*c*) Lefevre, cited by Hill, L.: The Science of Ventilation and Open Air Treatment, London, His Majesty's Stationery Office, 1919. Weyrich.[1d]

The relative humidity of the expired air is influenced by the temperature and relative humidity of the inspired air. This fact has been commented on and is illustrated by figure 5. When cool dry or cool foggy air was inspired, the mean relative humidity of the expired air was 86.1 per cent (extremes 83 and 94 per cent) or 83 per cent (extremes 79 and 91 per cent, respectively). When hot dry and hot moist air was inspired, the mean relative humidity of the expired air was 75.8 per cent (extremes 60 and 91 per cent) or 74.4 per cent (extremes 71 and 83 per cent, respectively).

IV. WATER LOSS AND METABOLIC RATE

For the 27 normal young resting adults a correlation coefficient of −0.26 was found between water loss from the respiratory tract and the rate of metabolism at that moment. Since metabolic rate is really a measurement of oxygen consumption while water loss is not necessarily dependent on oxygen consumption but (under comfortable environmental conditions) on the rate of irrigation of the respiratory tract with air, a correlation coefficient such as −0.26 might be expected. This means that more air circulated through the lungs than was necessary to meet the demands for oxygen. In normal subjects only the necessary amount of oxygen is removed from the air inspired. Emotional unrest, for example, might increase the rate of respiration beyond that necessary for the metabolic requirements. It is possible that such a factor was active in these studies.

That more air circulated through the lungs than was necessary for metabolic or oxygen needs is evidenced by the fact that a correlation coefficient of only +0.07 was found when the metabolic rate was correlated with the rate of irrigation of the respiratory tract with air. It must be remembered that the subjects studied were not in a basal metabolic state.

V. HEAT LOSS FROM THE RESPIRATORY TRACT

In 27 normal young white resting adults (ages varying from 17 to 36, mean 24 years) the heat loss from the respiratory tract was measured as outlined in a previous paper.[5] The room air, or inspired air, had a mean temperature of 20.5 C. (range 19.5 and 21.4 C.) and a mean relative humidity of 57.2 per cent (range 54 and 61 per cent). The heat loss was measured for the three components simultaneously (fig. 6): (1) heat loss by the evaporation of water, h_E, (2) heat loss by convection (warming of inspired air), h_c, and (3) heat loss by decomposition of carbonic acid and expiration of carbon dioxide

gas, h_{CO_2}. The total heat loss from the respiratory tract, H, was determined by the summation of these three components.

The total heat loss from the body was estimated by means of a Benedict-Roth type of B.M.R. machine.[5] Since the subjects had been resting in the observation room for over thirty minutes, it was assumed that there was thermal equilibrium between the subject's body and the environment. From this value for total body heat loss it was then possible to express heat loss from the respiratory tract (total or any of the three components) as per cent of total heat loss from the entire body.

1. *The Rate of Heat Loss by the Evaporation of Water (h_E).*—The heat loss by the evaporation of water varied between 0.305 and 0.706

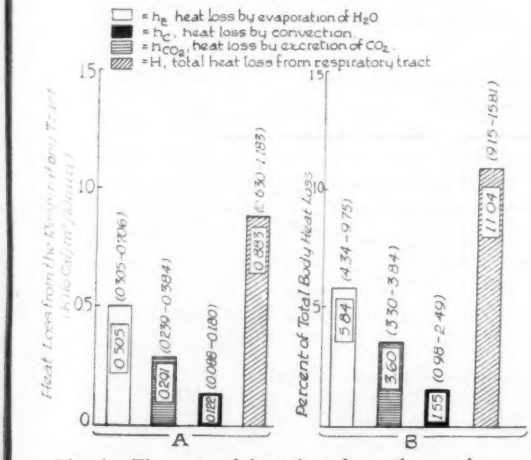


Fig. 6.—The rate of heat loss from the respiratory tract, showing its various components.

kilogram calories per square meter per ten minutes.[11] The statistical constants found were:

Mean, 0.505 ± 0.017 kilogram calories per square meter per ten minutes

Standard deviation, 0.191 ± 0.012 kilogram calories per square meter per ten minutes

Coefficient of variations, 37.90 ± 2.49 per cent

The relative importance of h_E in total pulmonary heat loss is shown in figure 6.

The mean rate of heat loss for the entire body was found to be 8.10 (extremes 6.62 and 10.77) kilogram calories. The heat loss from the respiratory tract by evaporation (h_E) represented a mean of 5.84 per cent of the total body heat loss (extremes 4.34 and 9.75 per cent). As an average, the h_E represented 53.2 per cent of H (total heat loss from the lungs).

Heat loss by evaporation varied with variations in environmental temperature and relative humidity (fig. 7). There were no significant variations in h_E when the atmosphere was changed from a cool dry to a cool wet and then to a comfortable environment. In a hot dry environment, h_E remained unchanged, but when the environment was hot and moist h_E decreased significantly (fig. 7). These changes are the result of the influences of these environmental conditions on the evaporation of water from the respiratory tract. Therefore, in a hot and humid environment heat loss by evaporation of water from the respiratory tract becomes much less significant, accounting for about 6 per cent of total heat loss in cool or comfortable environments. In a hot atmosphere saturated with water h_E would be 0.

Because of the high correlation between rate of water loss and rate of irrigation of the respiratory tract with air, the same high type of correlation would hold for h_E and rate of irrigation of the respiratory tract with air. In fact, the value of the correlation coefficient would be the same.

The importance of h_E, as in thermal equilibrium, depends on the conditions of the environment, especially in a subtropical and tropical climate.

2. *The Rate of Heat Loss by Warming Inspired Air (Convection, h_c).*—The heat loss by the warming of inspired air in 27 normal young adults sitting in a comfortable environment (temperature 21 C. and humidity 52 per cent) is shown graphically in figure 6. The relative importance of this factor in total pulmonary heat loss is indicated by figure 6. The statistical constants found for h_c were:

Mean, 0.122 ± 0.004 kilogram calories per square meter per ten minutes

Range, 0.088 and 0.180 kilogram calories per square meter per ten minutes

Standard deviation, 0.029 ± 0.003 kilogram calories per square meter per ten minutes

Coefficient of variation, 23.77 ± 2.31 per cent

It can be seen that h_c averaged about 13.9 per cent of the total heat loss from the lungs (H) and averaged 1.55 per cent of the total body heat loss under the comfortable environmental conditions.

When the temperature and the relative humidity of the room air were changed, the value for h_c varied (fig. 7). There were relatively little or insignificant differences in h_c in cool dry, cool foggy or comfortable environments. This lack of difference is due to

11. All values for heat loss are expressed in kilogram calories per square meter of surface area of the body per ten minutes. This unit will not be repeated; only the numerical values will be indicated.

the compensating influence of the rate of irrigation of the respiratory tract with air. The value of h_c would tend to be higher when cool air was inspired, but when cool air was inspired the amount of air inspired per unit of time was less than in the comfortable atmosphere. There was a definite change in h_c when the room air was raised above body temperature (fig. 7). Instead of losing heat by convection, the body gained heat from the atmosphere, h_c becoming a positive value. Heat loss by convection was affected relatively little by the moisture in the air other than through its effect on the density and specific heat of the expired air.

At constant atmospheric conditions, because of the nature of the calculation of h_c,[5] a high correlation between h_c and rate of irrigation of the respiratory tract with air would naturally

Standard deviation, 0.065 ± 0.006 kilogram calories per square meter per ten minutes

Coefficient of variation, 22.34 ± 2.15 per cent

Figure 6 shows the relative significance of h_{CO_2} when compared with other factors concerned with pulmonary loss of heat. The mean loss of heat by h_{CO_2} was 33.0 per cent of the total heat loss from the respiratory tract and averaged 3.60 per cent of the total body heat loss.

The value of h_{CO_2} is not influenced a great deal by the condition of the atmosphere (fig. 7) other than those concerned with changes in the rate of irrigation of the respiratory tract with air ("washing" out of carbon dioxide from the blood) and changes in the metabolic rate. In a hot and humid environment the metabolic rate and rate of irrigation of the respiratory tract

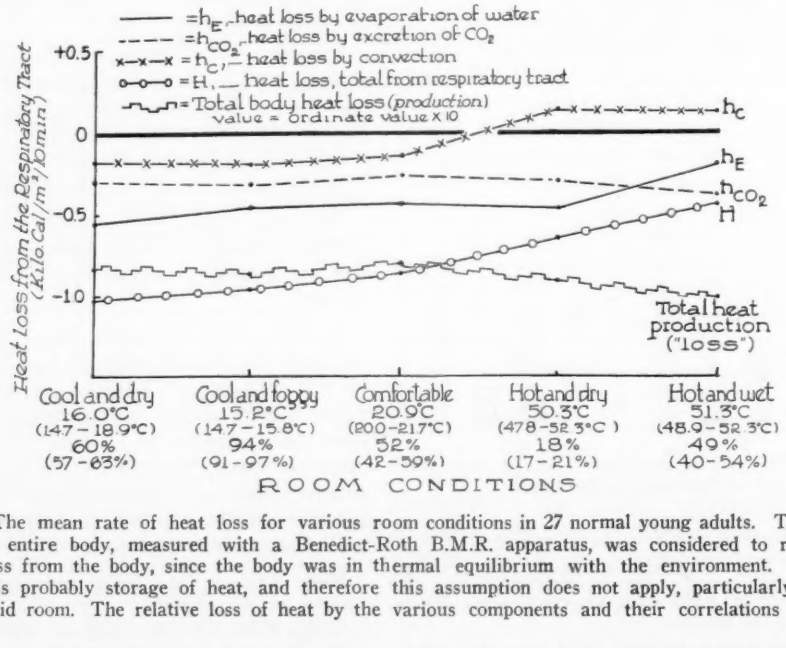

=hE, heat loss by evaporation of water
=hCO2, heat loss by excretion of CO2
=hC, heat loss by convection
=H, heat loss, total from respiratory tract
=Total body heat loss (production)
value = ordinate value x 10
Heat Loss from the Respiratory Tract (Kilo Cal/m²/10 min)
+0.5
0
-0.5
-1.0
hC
hE
hCO2
H
Total heat production ("loss")
Cool and dry 16.0°C (14.7-18.9°C) 60% (57-63%)
Cool and foggy 15.2°C (14.7-15.8°C) 94% (91-97%)
Comfortable 20.9°C (20.0-21.7°C) 52% (42-59%)
Hot and dry 50.3°C (47.8-52.3°C) 18% (17-21%)
Hot and wet 51.3°C (48.9-52.3°C) 49% (40-54%)
ROOM CONDITIONS

Fig. 7.—The mean rate of heat loss for various room conditions in 27 normal young adults. The total heat loss for the entire body, measured with a Benedict-Roth B.M.R. apparatus, was considered to represent the total heat loss from the body, since the body was in thermal equilibrium with the environment. In the hot room there is probably storage of heat, and therefore this assumption does not apply, particularly not to the hot and humid room. The relative loss of heat by the various components and their correlations are evident.

follow. Although h_c is not large, it can become important under certain conditions of low or high atmospheric temperatures (for example, subtropical and tropical climates).

3. *The Rate of Heat Loss by the Excretion of Carbon Dioxide (h_{CO_2}).*—Simultaneously with the measurement of the foregoing two types of heat loss in the 27 normal subjects sitting in a comfortable environment, the heat loss by the expiration of carbon dioxide was measured. The statistical constants found for h_{CO_2} are:

Mean, 0.291 ± 0.008 kilogram calories per square meter per ten minutes

Range, 0.239 to 0.384 kilogram colories per square meter per ten minutes

with air are increased (fig. 5). These were found to be associated with an expected increase in h_{CO_2}.

Judging by the nature of the method for measuring h_{CO_2}, a high correlation of h_{CO_2} with the metabolic rate would be expected.[5] The importance of h_{CO_2} in thermal equilibrium in man in a tropical and subtropical climate, though relatively small, is self evident from these comments.

4. *The Rate of Total Heat Loss from the Respiratory Tract (H).*—The total heat loss from the respiratory tract is

$$H = h_E + h_c + h_{CO_2}$$

The relative values of H and its three components are shown in figure 6.

In the study of the 27 normal young adults sitting in a comfortable atmosphere, H was found to have the following statistical constants:

Mean, 0.886 ± 0.034 kilogram calories per square meter per ten minutes
Range, 0.630 to 1.183 kilogram calories per square meter per ten minutes
Standard deviation, 0.264 ± 0.024 kilogram calories per square meter per ten minutes
Coefficient of variation, 29.80 ± 2.98 per cent

The total heat loss from the respiratory tract (H) averaged 11.04 per cent of the total body heat loss, the extremes being 9.15 and 15.181

The total heat loss from the respiratory tract varied with the temperature and humidity of the environment (fig. 7). In a cool environment with a low or high relative humidity, H changed relatively little. In a hot environment with the temperature greater than body temperature, H decreased significantly. This decrease was especially great in a hot and humid atmosphere (fig. 7).

Although the rate of total heat loss from the respiratory tract decreased in the hot and humid environment, the rate of heat production increased noticeably. The decrease in H in the hot and humid atmosphere is due mainly to the decrease in h_E and the change in h_C. Under those atmospehric conditions h_c becomes a positive rather than a negative value. Instead of body heat being lost by convection, it is being increased by this factor. The significance of these phenomena in thermal regulation in a tropical and subtropical climate is evident from figure 7.

VI. THE RATE OF PRODUCTION AND LOSS OF TOTAL BODY HEAT

The rate of total heat production for the body as a whole was measured with a Benedict-Roth B.M.R. machine for the 27 normal young adults sitting quietly in a comfortable environment. Since the environment was comfortable and the subjects had rested for a period of thirty minutes or more prior to the measurements, the subjects were considered to be in thermal equilibrium with their environment and therefore the rate of heat production and heat loss were considered equal. The statistical constants found for total heat loss from the body are:

Mean, 8.08 ± 0.19 kilogram calories per square meter per ten minutes
Range, 6.62 to 10.77 kilogram calories per square meter per ten minutes
Standard deviation, 1.50 ± 0.14 kilogram calories per square meter per ten minutes
Coefficient of variation 1.851 ± 1.75 per cent

In a hot and dry environment the rate of total body heat production increased. When the humidity of the hot atmosphere was further increased, the rate of total body heat production also increased (fig. 7). When the temperature of the atmosphere was greater than that of the body (50 C.), thermal equilibrium between the subject and his surroundings did not occur, and therefore the rate of heat production was not equal to heat loss. The exact nature of the state of transfer of heat between the subject as a whole and the environment was not known during these studies, especially when the room air was hot. In spite of this, the rate of total body heat production was intentionally, although erroneonusly, considered equal to the rate of total loss of body heat to make it possible to obtain an idea of the part played by the respiratory tract in the elimination of body heat at the high room temperatures.

VII. STUDIES OF THE RATE OF WATER AND HEAT LOSS FROM THE RESPIRATORY TRACT OF ONE SUBJECT

One subject, a young (34 years of age) white man sitting quietly, was studied at frequent intervals for seven consecutive hours during one day (beginning at 10 a. m.). The rate of water loss, rate of heat loss from the respiratory tract (the three components and total), rate of total heat loss from the entire body, temperature of the expired air, rate of irrigation of the respiratory tract with air and relative humidity of the expired air were among the factors recorded (fig. 8). The room conditions are indicated by figure 8. The physiologic phenomena recorded were remarkably constant, the main change occurring shortly after a moderately heavy noon meal (fig. 8).

The same subject was similarly studied several times over a period of months, extending from the hot summer month of August to the cool month of January. The results are summarized for the rate of water loss in figure 9. The conditions of the room were the same, being comfortable and having values as indicated for that type of room in figure 8. There were no significant differences from month to month as the season changed from summer to winter in New Orleans. There were fluctuations from month to month, but they were considered to be ordinary physiologic variations.

COMMENT

It is apparent that the expired air is not saturated with water and could not possibly be so.[8] As found in these studies, it is only about 88 per cent saturated when the subject is breathing air

in a comfortable environment as well as when he is breathing cool dry or foggy air. This conclusion is in keeping with the recent studies of Seeley [8] and Christie and Loomis.[4] It was also found many years ago by Loewy and Gerhartz.[1h] It is well to note that Galeotti [1g] found the expired air to be 78 per cent saturated at 37 C. and completely saturated at a temperature of 32.5 C. The temperatures of the expired air were assumed in his studies, thus rendering his statements of relative humidity of expired air of only academic interest. Galeotti [1f] at a later date measured the temperature of the expired air with a thermocouple and found it to vary between 34.4 and 35.7 C. Liljestrand and Sahlstedt [12] found the expired air to have a 98 per cent saturation that expired air is saturated at 37 C. As was shown, the expired air is neither saturated nor at 37 C. when subjects are in a comfortable atmosphere.

The majority of observers [14] found the temperature of the expired air to be about 32 to 33 C. The values observed in these studies are in keeping with these figures rather than with some of the higher values for a comfortable atmosphere. The mean temperature of the expired air with the subject resting quietly was found to be 33.2 C., with extremes of 31.6 and 34.2 C.

When the temperature and the relative humidity of the atmosphere are changed, the nature of the expired air changes. The tempera-

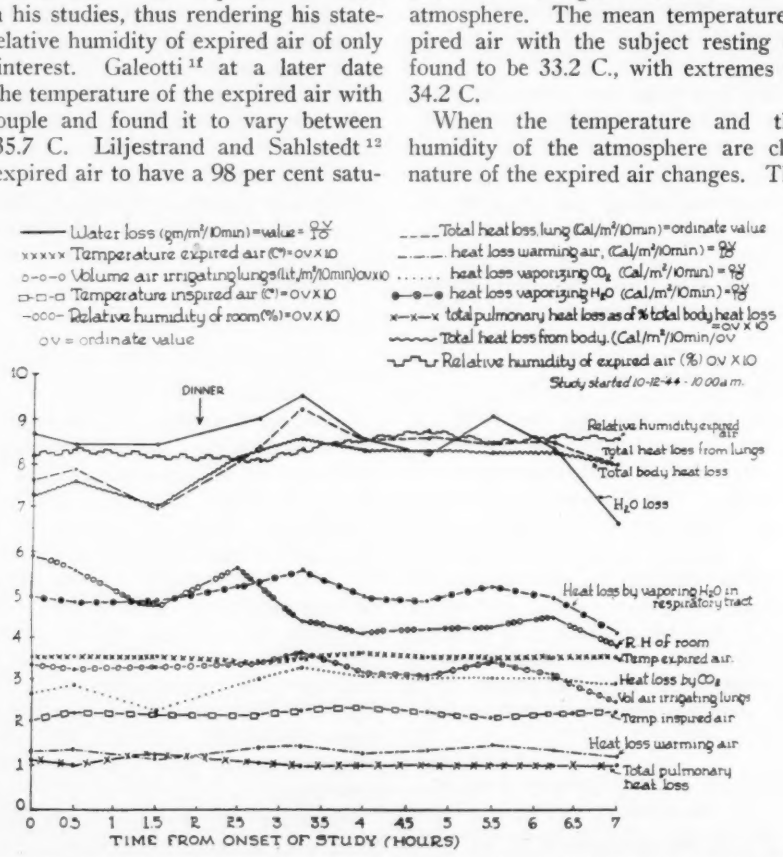

Water loss (gm/m²/10min) = value ÷ 10
Temperature expired air (C°) = OV x 10
Volume air irrigating lungs (lit./m²/10min) OV x 10
Temperature inspired air (C°) = OV x 10
Relative humidity of room (%) = OV x 10
OV = ordinate value
Total heat loss lung (Cal/m²/10min) = ordinate value
heat loss warming air, (Cal/m²/10min) = ÷ 10
heat loss vaporizing CO₂ (Cal/m²/10min) = ÷ 10
heat loss vaporizing H₂O (Cal/m²/10min) = ÷ 10
total pulmonary heat loss as of % total body heat loss = OV x 10
Total heat loss from body. (Cal/m²/10min/OV
Relative humidity of expired air (%) OV x 10
Study started 10-12-44 - 10.00 a.m.
DINNER
Relative humidity expired air
Total heat loss from lungs
Total body heat loss
H₂O loss
Heat loss by vaporing H₂O in respiratory tract
R. H of room
Temp expired air
Heat loss by CO₂
Vol air irrigating lungs
Temp inspired air
Heat loss warming air
Total pulmonary heat loss
TIME FROM ONSET OF STUDY (HOURS)

Fig. 8.—A study of the rates of water loss and heat loss from the respiratory tract of a normal young man made over a seven hour period (beginning at 10 a.m.). All factors noted were recorded at frequent intervals, that is, at the times indicated by the abscissa.

ration at a temperature of 30 to 35 C. The statements of Weyrich,[1d] Luciani,[10b] Lefevre [10c] and Osborne [10a] that expired air is saturated with water were erroneous. Benedict and Carpenter [13] employed the method of Zuntz to calculate water loss from the lungs. These calculations are in error, since they were based on the assumpture and relative humidity of the expired air does not change significantly when the atmospheric conditions are made cool and dry or cool and wet. As just mentioned, cool air, whether wet or dry, shows little difference in the absolute amount (grams) of water vapor per unit volume, although the relative humidity may vary considerably. When the atmospheric temperature is elevated considerably, the capacity of the air to hold water is increased considerably, so that relatively small differences in relative humidity mean great differ-

12. Liljestrand, G., and Sahlstedt, A. V.: Temperatur und Feuchtigkeit der Ausgeatmeter Luft, Skandinav. Arch. f. Physiol. **46**:94, 1924.

13. Benedict, F. G., and Carpenter, T. M.: The Metabolism and Energy Transformations of Healthy Man During Rest, Washington, D. C., Carnegie Institution of Washington, 1910.

14. Galeotti.[1f] Galeotti.[1g] Loewy and Gerhartz.[1h] Seeley.[8] Loewy and Gerhartz.[1b]

ences in absolute humidity. These great influences of relative humidity of hot air on the rate of water loss (and also heat loss by evaporation of water) from the respiratory tract were shown by these observations. At a room temperature of 50 C. and at 18 per cent relative humidity, the rate of water loss (heat loss by evaporation also) from the respiratory tract is increased over that at conditions in a comfortable environment. In an atmosphere of 50 C. and 49 per cent relative humidity, the rate of water loss is noticeably decreased. The same is not true for a cool atmosphere. The rate of water loss for an atmospheric temperature of 15 or 16 C., whether 60 or 97 per cent saturated, was essentially the same. It is apparent that expressions of the moisture content of the atmosphere in vapor pressure or absolute humidity rather than relative humidity would give a much clearer concept of the physical factors concerned with water loss from the respiratory tract.

It is interesting to note that the surface area of the respiratory tract has been estimated to be

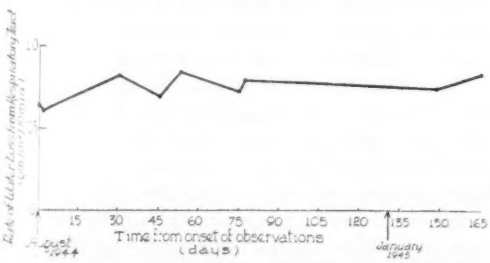


Fig. 9.—The rate of water loss from the respiratory tract over a period of several months in a normal young man sitting in a comfortable environment (temperature 20.9 ± 0.9 C.).

about 70 square meters during quiet breathing and 90 square meters when moderately inflated. That of the alveoli alone is about 55 square meters.[15] Some estimates as high as 129 square meters have been made. Pulmonary area over thirty times that of the external (skin) area of the body is truly large and therefore an important factor concerned with water and heat loss from the respiratory tract.

The role of the respiratory tract in the maintenance of thermal equilibrium between heat produced in the body and that lost to the surrounding atmosphere is significant. In a normal adult sitting in a comfortable environment, the total heat loss from the respiratory tract accounts for about 11 per cent of the total heat loss from the body. This percentage is great enough to warrant serious consideration. The importance of this factor is even more evident when it is considered that the habits of man today render it impossible to vary this loss even under extreme conditions of environmental temperature. By the use of clothing it is possible to influence the amount of heat lost from the external, or cutaneous, surface of the body. In the case of the respiratory tract no such simple procedures for varying heat loss are available. In a cool or uncomfortably cold environment, the heat loss from the respiratory tract remains relatively unchanged except for the factor h (warming of cold inspired air). Considering the surface area of the respiratory tract and the influence of cold on blood vessels, glandular function and the like of the entire respiratory tract, it is indeed not surprising to note a great tendency for infections of the respiratory tract to develop in the cold months of the year. Among persons exposed to cold environments because of occupational, military or other reasons for prolonged periods of time, the role of the respiratory tract in heat loss is important enough to demand more consideration than it usually commands.

In a hot and humid environment, when heat loss from the skin is definitely impaired, the heat loss from the respiratory tract likewise is impaired. The loss of heat by h_{CO_2} remains unchanged or may increase with the increase in excretion of carbon dioxide (Bazett[16]). When the temperature of the environment is greater than body temperature, h_c becomes a positive value, and heat is absorbed from the surrounding atmosphere. The high humidity reduces heat loss by the evaporation of water. Therefore the respiratory tract is of little value in the elimination of body heat in a hot and humid environment. As indicated previously, if the environmental temperature is sufficiently high, the relative humidity does not have to be very high in order to effect a much higher effective relative humidity. For when hot air (50 C.) only about 50 per cent saturated becomes about 80 per cent saturated on being cooled to 39 C. by the respiratory tract, the resulting high relative humidity of hot air after inspiration is considerably different from the relative humidity of the room air.

It is frequently stated that a subject inspiring foggy air will gain water from the atmosphere by a deposition of many small droplets of water on the surface of the epithelium of the respiratory tract. That this may be erroneous, unless large droplets of water form the fog, is indicated by

15. Willson, H. G.: The Terminals of the Human Bronchioles, Am. J. Anat. **30**:267, 1922.

16. Bazett, H. C.: Physiological Responses to Heat, Physiol. Rev. **7**:531, 1927.

the aforementioned observations. When a subject inspires air at 15 C. and 97 per cent relative humidity (cool foggy air), the temperature of the air is raised to about 33 C., and it is then no longer foggy, is far from saturated and is capable of readily taking on a great deal more water. It does remove water from the epithelium of the respiratory tract. Water is not absorbed from the atmosphere but is readily lost to it.

To fulfil the metabolic requirements, a certain amount of oxygen must be absorbed from the atmosphere and a certain amount of carbon dioxide excreted into it. The quantity of carbon dioxide excreted will be influenced less by the temperature and humidity of the air inspired than the water vaporized or heat lost by convection. Therefore, heat loss by excretion of carbon dioxide remains relatively constant in various atmospheric conditions, all other factors being constant.

The lack of correlation of the metabolic rate (oxygen consumption) and rate of irrigation of the respiratory tract with air indicates that the rate of circulation of air through the lungs is not governed only by the oxygen requirements. This physiologic fact is well known. The dog, for instance, increases the rate of breathing when in a hot environment. This increase may occur to some extent in man; whether or not this is concerned to any extent with cooling remains unknown.[16] However, as would be expected, there is a high positive correlation of rate of irrigation of the lungs with air and the rate of water loss from the lungs.

Boyer and Bailey[17] found a fair correlation ($+ 0.58 \pm 0.02$) between surface area of the body and the ventilation of the lungs (liters per minute) and a high correlation between ventilation and heat production in normal subjects in the basal state. In the latter correlation, no corrections were made for body size. Such a correction, if made in units of surface area, would have reduced the value of the correlation coefficient of $+ 0.90$ to something approaching $+ 0.58$. This factor explains in part the lack, in our studies, of a high correlation between the rate of irrigation of the lungs and metabolic rate. The differences in time of measurements of the metabolic rates and the rate of irrigation of the lungs, position of the subject and the subject's resting, but definitely not basal, state are among the factors which are responsible for differences between the findings reported here and those of Boyer and Bailey.[17]

17. Boyer, P. K., and Bailey, C. V.: Concentration of Carbon Dioxide in Expired Air, Arch. Int. Med. **69**:773 (May) 1942.

SUMMARY AND CONCLUSIONS

The rates of water loss and heat loss from the respiratory tracts of 107 young normal resting (sitting) adults were studied. These subjects lived in a subtropical climate (New Orleans). They varied in age from 17 to 43 years and represented both sexes and the white and Negro races.

A comfortable environment of reference (temperature 20 to 21.1 C. and relative humidity 55 to 60 per cent) was used in all groups of measurements.

For 56 young normal adults sitting in the comfortable room and studied during the hot month of August 1944, the mean rate of water loss from the respiratory tract was 0.878 Gm. per square meter of surface area of the body per ten minutes, the extremes being 0.527 and 1.172 Gm. Twenty-seven similar subjects (some were in the group of 56) observed under similar conditions but studied during the cool month of January had a mean rate of water loss from the respiratory tract of 0.804 Gm. per square meter per ten minutes, the extremes being 0.508 and 1.205 Gm. Thus no seasonal difference was noted. There were no differences due to sex or race.

The mean rate of irrigation of the lungs for the 56 young normal subjects was 38.326 liters per square meter per ten minutes, the range being 28.316 and 53.250 liters. There was an extremely high positive correlation between the rate of irrigation of the lungs and the rate of water loss from the respiratory tract, the coefficient of correlation being $+ 0.914 \pm 0.015$.

Exercise increased the rate of water loss from the respiratory tract essentially in proportion to the resultant increase in the rate of irrigation of the respiratory tract with air.

There was no significant difference between the results obtained for water loss whether the subject respired through the nose or mouth.

The depth and rate of respiration influenced the rate of water loss from the respiratory tract mainly on the basis of the rate of irrigation of the respiratory tract with air. Slow deep breathing was, however, associated with more water loss per unit of volume of air respired than was rapid shallow breathing. In the former type of breathing the air remained in the respiratory tract a longer time, thus permitting more water vapor to diffuse into the air inspired.

The rate of water loss from the lungs was changed little from that in a comfortable environment when the environmental temperature was cooled to about 15 C. At that temperature the relative humidity of the air, even saturated to the extent of producing fog, changed the rate of water

loss from the respiratory tract relatively little. Some of the mechanisms involved in the lack of differences were studied.

The rate of water loss was increased by a hot (50 C.) and dry (18 per cent relative humidity) atmosphere. It was noticeably decreased when the hot (50 C.) atmosphere was moistened (49 per cent saturated) but not nearly saturated. The mechanisms involved were studied.

The mean temperature of the expired air with the subjects resting in the comfortable atmosphere was 33.18 ± 0.21 C. A drop in the environmental temperature to about 20 C. did not influence the temperature of the expired air. An increase in the temperature of the atmosphere to about 35.5 C. had no significant influence on the temperature of the expired air. When the temperature of the atmosphere was increased above 35.5 C., there was a definite rise in the temperature of the air expired.

The mean relative humidity of expired air was found to be 88.15 ± 1.31 per cent, the extremes being 78 and 96.5 per cent. The relative humidity of the air expired varied with that of the air inspired, especially if the inspired air was hot. Hot and relatively dry atmospheric air reduced the degree of saturation of expired air.

There was no significant correlation between the rate of water loss and the metabolic rate. This lack of correlation was to be expected, since the irrigation of the lungs is not determined by oxygen requirements only.

The rates of total heat loss from the respiratory tract and of its three components were determined simultaneously. Heat is lost from the respiratory tract by evaporation of water (h_E). In 27 normal young adults sitting in a comfortable environment the mean value for h_E was 0.505 ± 0.017 kilogram calories per square meter per ten minutes, the extremes being 0.305 and 0.706 kilogram calories. Heat loss by evaporation is not changed much by a cool dry or cool foggy atmosphere. It is increased by a hot (50 C.) and dry (18 per cent) atmosphere and definitely decreased when the relative humidity of the hot (50 C.) atmospheric air is increased (49 per cent). The mechanisms involved were studied.

Heat is lost from the respiratory tract by warming cool inspired air (convection, or h_c. The mean value for h_c with the environment comfortable was 0.122 ± 0.004 kilogram calories per square meter per ten minutes, the extremes being 0.088 and 0.180 kilogram calories. Heat loss by convection was not changed by changing the atmosphere to a cold dry or foggy one. When the temperature of the environment was raised to 50 C. (relative humidity 18 or 49 per cent), h_c became a positive value; that is, heat was absorbed from the hot inspired air, thus tending to increase body temperature.

Heat is lost from the respiratory tract because of the excretion of carbon dioxide (h_{CO_2}). Under comfortable atmospheric conditions, the mean rate of heat loss by the excretion of carbon dioxide was 0.291 ± 0.008 kilogram calories per square meter per ten minutes, the extremes being 0.239 and 0.384 kilogram calories.

The value of h_{CO_2} is influenced only by the rate of excretion of carbon dioxide and therefore was influenced by the atmospheric temperature and relative humidity only so far as these factors influenced the rate of excretion of carbon dioxide.

The total heat loss from the respiratory tract (H) is the sum of $h_E + h_c + h_{CO_2}$. From these components the mean rate of total heat loss from the respiratory tract was 0.886 ± 0.037 kilogram calories per square meter per ten minutes, the extremes being 0.630 and 1.183 kilogram calories. The nature of the influence of H by the temperature and relative humidity is determined by the nature of the influence of these factors on the three components.

In the 27 normal young resting adults the total rate of heat production and, therefore, heat loss if the subject is in thermal equilibrium with his environment, was measured along with the heat loss from the respiratory tract. In a comfortable environment the mean rate of total body heat loss was 8.08 ± 0.19 kilogram calories per square meter per ten minutes, the extreme being 6.62 and 10.77 kilogram calories. A hot dry or hot humid environment increased this value.

A single subject sitting in a comfortable environment and studied at frequent short intervals during seven hours of one day and at intervals of several days over a period of five and one-half months showed a remarkable constancy in the temperature and relative humidity of the expired air, rate of water and heat loss (H, h_E, h_c and h_{CO_2}) from the respiratory tract, rate of total body heat loss and rate of irrigation of the respiratory tract with air. A heavy meal changed these values slightly. They were not significantly different during the hot summer or cool winter of subtropical New Orleans.

It is impossible to state definitely the nature of the expired air or water or heat loss from the respiratory tract without first defining at least the conditions of the air inspired, the type of respiration and the activity of the subject. These other factors influence the expired air considerably.

Mr. G. Morgavi assisted in these studies.

SOME OBSERVATIONS ON PRIMARY ATYPICAL PNEUMONIA

CAPTAIN JACOB GROSSMAN
MEDICAL CORPS, ARMY OF THE UNITED STATES

Studies on primary atypical pneumonia, especially those of the past four years, have resulted in the awareness of a rather well defined clinical picture in this disease. Several authors, however, have presented series of cases differing considerably in their symptomatology. It is the purpose of this paper to describe a group of cases of the disease appearing during the first half of 1944 in Italy and to compare them with cases previously reported. A large number of the patients were evacuated from Anzio, and, in a sense, the outbreak may be regarded as localized. This series consists of 45 patients whose uniformity of symptoms is especially interesting as a form which the disease may take. Conditions governing the evacuation of casualties render it virtually impossible for one to reach any conclusions concerning epidemiology. It is my impression from conversation with patients that successive groups of persons in particular units were affected and that, at least under combat conditions involving exposure, chilling, fatigue and lowered resistance, the disease is apparently infectious, with a shorter incubation period than has been generally stated. This belief is strengthened by the reported higher incidence of the disease among station hospital personnel [1] as compared to that among other troops, the localized outbreaks in small units [2] and several small epidemics in hospitals.[3]

Clinical Observations.—Almost invariably, the disease was well defined in onset. Only 5 patients admitted premonitory symptoms. Of these, 3 had "colds" and slight coughs of several days' duration; 1 complained of generalized aching and malaise for one day, and 1 stated he had been unwell for three to four weeks. However, in these patients as well as in the others, the onset of more severe disease was clearcut. This was marked by sudden chilly sensations and fever in two thirds of the patients, accompanied by severe frontal headache. Not only was the last mentioned the most frequent early symptom, occurring in 78 per cent of the patients, but by its severity tended to draw attention away from symptoms referable to the chest, which early in the disease frequently showed no abnormalities on physical examination. Generalized muscular pains, most pronounced in the lower part of the back and the legs, were present at the outset in about half of the patients, a number of whom became aware of the pains a few hours prior to the chills and fever. An unusually high number (18, or 40 per cent) complained of pain in the chest at the onset. Unlike pains described in previous reports, which noted little pleural involvement,[4] in 13 of these patients the pain was unilateral and increased with inspiration. Only 5 described it simply as a soreness. Ten patients (22 per cent) mentioned coughing at the onset, at which time the cough was described as "slight" or "dry." Weakness, malaise, drowsiness, anorexia and nausea were present in variable degree in almost all the patients.

The course of the disease may be divided into two parts—an acute phase of five to seven days, until the temperature subsides, and a considerably longer recuperative phase lasting four to six weeks, during which the pulmonary tissue reverts to normal. During the first few days of the disease, there was little change in the picture. Repeated chilly sensations or chills, more frequently the former, continued irregularly for several days and invariably appeared, with fever, in those patients in whom they were not present at the onset. The temperature was of the irregularly elevated type, with wide daily variations at

1. Dingle, J. H., and others: Primary Atypical Pneumonia, Etiology Unknown, War Med. **3**:223 (March) 1943.

2. Idstrom, L. G., and Rosenberg, B.: Bull. U. S. Army M. Dept., October 1944, no. 81, p. 88.

3. (*a*) Maxfield, J. R., Jr.: Texas State J. Med. **35**:340 (Sept.) 1939. (*b*) Dingle, J. H., and Finland, M.: New England J. Med. **227**:378 (Sept. 3) 1942. (*c*) Favour, C. B.: New England J. Med. **230**:537 (May 4) 1944.

4. (*a*) Campbell, T. A.; Strong, P. S.; Grier, G. S., III, and Lutz, R. J.: Primary Atypical Pneumonia: Report of 200 Cases at Fort Eustis, Virginia, J. A. M. A. **122**:723 (July 10) 1943. (*b*) Duggan, L. B., and Powers, W. L.: U. S. Nav. M. Bull. **40**:651 (July) 1942. (*c*) Young, H. E.; Storey, M., and Redmond, A. J.: Am. J. M. Sc. **206**:756 (Dec.) 1943. (*d*) Primary Atypical Pneumonia, Etiology Unknown, War Med. **2**:330 (March) 1942. (*e*) Whiteley, J. H.; Bernstein, A., and Goldman, M. J.: Mil. Surgeon **91**: 499 (Nov.) 1942. (*f*) Kneeland, Y., Jr., and Smetana, H. F.: Bull. Johns Hopkins Hosp. **67**:229 (Oct.) 1940. (*g*) Green, D. M., and Eldridge, F. G.: Mil. Surgeon **91**:503 (Nov.) 1942.

times. As a rule, the higher temperature continued for five to seven days and then fell by lysis over a three day period. Chart 1 indicates the frequency distribution of the duration of fever. Coughing was prominent and occasionally distressing in the first days of the disease. In addition to the 10 patients who complained of cough at the onset, 11 (24 per cent) began to cough on the second day, 7 on the third day and a few still later. As a rule, the cough was not severe and was either dry or productive of a small quantity of mucoid sputum at first, either subsiding or becoming more productive at about the time the temperature fell to normal. Seven patients (16 per cent) brought up a slightly blood-streaked sputum; in no case was the hemoptysis severe.

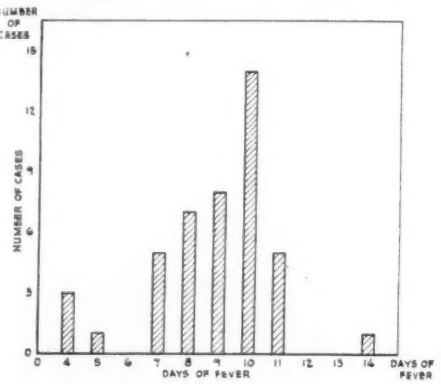


Chart 1.—Duration of fever.

The headache, after its initial severity, was variable both in intensity and in duration, lasting one to four days. The aggravation of headache by coughing suggested a relation to increased pressure of the cerebrospinal fluid in accordance with the observations of Dingle and his associates,[1] who relieved the headache in 7 patients by lumbar puncture. Similarly, the pain on breathing deeply tended to be transient, subsiding shortly. Other common systemic symptoms which occurred during the acute phase of the disease and paralleled its severity were weakness with lassitude in 17 patients (38 per cent) and anorexia (20 per cent). Seven patients vomited; 2 had seemingly spontaneous epistaxis.

Physical signs were neither prominent nor of specific diagnostic value. At the onset, patients were drowsy, with warm, flushed skin, either dry or covered by profuse perspiration. Cyanosis was uncommon, was rarely discernible and was evident in only 1 patient, who might have had an associated bacterial pneumonia. Respiration was often difficult and shallow, owing to pleural involvement, but there was neither tachypnea, as seen in bacterial pneumonia, nor true dyspnea. The slightly increased respiratory rate gradually decreased as the acute picture subsided. A relative bradycardia was a constant feature during the acute phase, but as the temperature fell the cardiac rate rose in about half the patients. Similar observations were recorded by Green and Eldridge [4g] and Young, Storey and Redmond.[4e] The cause of this phenomenon is not known but may be related to the absorption of pathologic products from the tissues of the lung. Somewhat bleary, injected conjunctivas were seen fairly frequently, and 10 patients had injected pharynges, though only 4 complained of a sore throat. Lymphadenopathy was not a feature of the disease; a palpable spleen was twice encountered. Loss of weight was common and was prominent in 5 cases. Moreover, another aspect of recovery was evident in the patients' difficulty in regaining lost weight even after the appetite had returned and caloric intake was increased.

The paucity of signs referable to the chest, as observed by others,[5] has been—in contrast to the pronounced roentgenographic changes—one of the most remarkable features of the disease. The explanation probably lies in the fact that the pathologic process is primarily interstitial rather than within the air passages or alveoli [6]; this fact may likewise account for the absence of cyanosis. Physical examination of the chest revealed nothing abnormal almost throughout the entire course in 18, or 40 per cent, of the patients. Even when present, the signs were slow to appear, rarely being perceptible until the third day. At this time, some fine moist subcrepitant rales were discerned, best heard toward the end of inspiration; a smaller number of patients had simply diminished or roughened breath sounds. The presence of an impaired percussion note in a few was, as a rule, associated with larger and denser lesions, as seen roentgenographically. After seven to ten days from the onset the rales became coarser, moister and noisier, and rhonchi were heard. Some wheezing, which also appeared at this stage, persisted for several weeks. Evidence of frank consolidation was uncommon.

5. (*a*) Longcope, W. T.: Practitioner **148**:1 (Jan.) 1942. (*b*) Rhoads, P. S.: Radiology **40**:327 (April) 1943. (*c*) Thompson, J. L., Jr.: M. Ann. District of Columbia **12**:171 (May) 1943. (*d*) Correll, H. L., and Cowan, I. I.: U. S. Nav. M. Bull. **41**:900 (July) 1943. (*e*) Lusk, F. B., and Lewis, E. K.: Dis. of Chest **10**:19 (Jan.) 1944. (*f*) Footnote.[4d] Duggan and Powers.[4b] Dingle and Finland.[5b] Green and Eldridge.[4g] Idstrom and Rosenberg.[2]

6. Golden, A.: Bull. U. S. Army M. Dept., October 1944, no. 81, p. 64.

Laboratory Observations.—From those laboratory procedures routinely performed, little of diagnostic value was found. The red blood cell count ranged between 4,000,000 and 5,000,000 per cubic millimeter, as did that of other patients. The white blood cell count during the acute phase of the disease was well scattered throughout the normal range of 5,000 to 9,000 per cubic millimeter. During the course of the disease, there was a perceptible increase in the white blood cell count, which on examination was seen to be due largely to an increase in lymphocytes between the fourth and sixth week (chart 2). The erythrocyte sedimentation rate appeared to be a reliable index of disease activity, its slow decrease correlating well with the gradual disappearance of roentgenologic evidence of disease. Though there is considerable over-all variation in the results obtained (chart 3) for any 1 patient, the relative sedimentation rate from time to time is a valuable indication of recovery, second only to the roentgenogram itself.

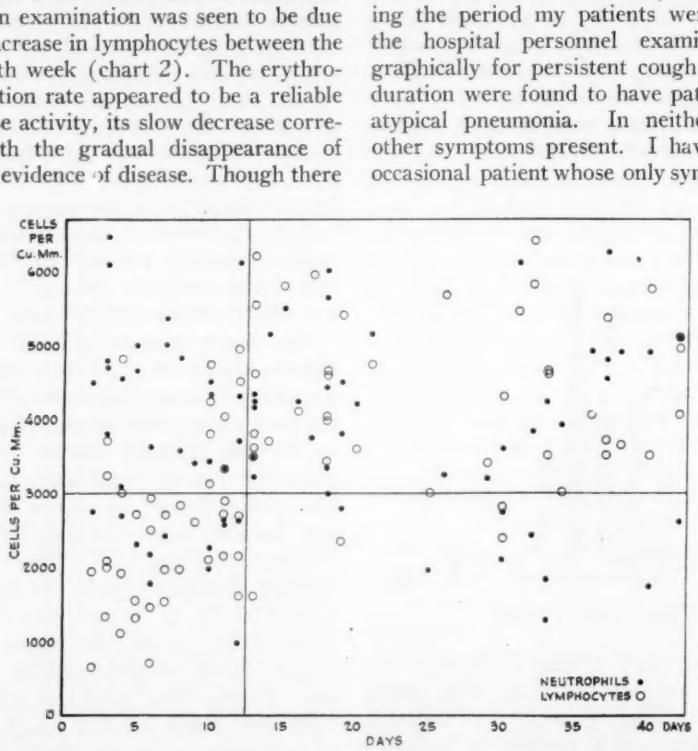


Chart 2.—Rise in number of lymphocytes in course of primary atypical pneumonia.

Except for a variable degree of albuminuria in 11 patients and the presence of an occasional granular cast in the urine of 3 patients early in the disease, examination of the urine showed nothing abnormal.

Several attempts to demonstrate the presence of cold agglutinins were unsuccessful.

Roentgenographic Observations.—In the lack of pathognomonic symptoms and with the dearth of physical signs in the chest, the roentgenogram assumes great importance in diagnosis. In fact, the principal criterion, almost the sine qua non, is the characteristic roentgenogram. The finding by Dingle and his associates [1] of 34 cases of "bronchitis resembling atypical pneumonia" (clinically) without roentgenographic changes does not detract from the value of the roentgenogram. The disease may occur in such forms that the roentgenogram is virtually the only evidence of its presence.[7] Fredd [7a] found its incidence to be 0.27 per cent of 23,465 roentgenograms of the chest taken of clinically well people. During the period my patients were studied, 2 of the hospital personnel examined roentgenographically for persistent cough of some weeks' duration were found to have patches of primary atypical pneumonia. In neither patient were other symptoms present. I have since seen an occasional patient whose only symptom was fever.

Roentgenologic evidence of disease as a rule did not appear until two or three days after the onset. There was first an increase in bronchial markings, more pronounced at the hilar areas, followed shortly by irregular patches of increased density farther peripherally. The bronchial markings were also the last evidences of the disease to disappear. These areas of increased density varied from a soft mottling which in the upper lobes resembled early pulmonary tuberculosis to a denser, hazy opacity or glazed appearance seen in pulmonary congestion and which has also been said to be due to inflammatory pleural changes.[4d] The density was not

7. (*a*) Fredd, H.: New York State J. Med. **41**:34 (Jan. 1) 1941. (*b*) Lusk and Lewis.[5e] Idstrom and Rosenberg.[2]

as pronounced as that seen in lobar pneumonia, again in consequence of the fact that the pathologic process is interstitial and does not produce a filling of the alveoli with dense cellular material. Resolution, in contrast to that of bacterial pneumonia, is prolonged, and increased bronchial markings and some parenchymal mottling may remain long after the patient is clinically well.

In over two thirds of the patients, only one patch of pneumonia could be detected on roentgenographic examination. Nine patients had two distinct areas of density and 1 patient three. Multiple areas were seen in 2 patients. The distribution of the lesions was similar to that described in previous reports,[8] with the lower lobes predominantly involved, as follows: right upper pulmonary field eleven times, left upper field six times, right middle and lower fields

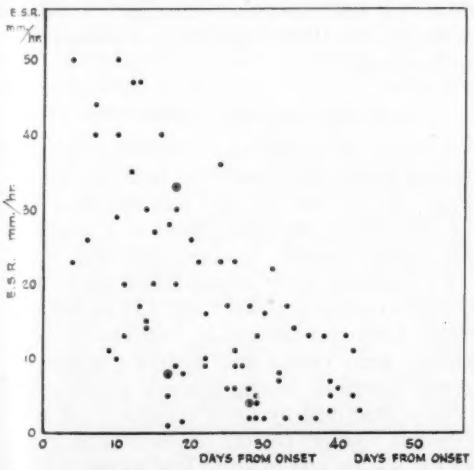


Chart 3.—Fall in erythrocyte sedimentation rate during course of primary atypical pneumonia.

seventeen times and left lower field twenty-one times. Where pulmonary tissue close to the interlobar pleura was involved, that pleura remained thickened, as mentioned by Bahlke;[9] this thickening was observed in 8 patients. It was my impression that when the upper pulmonary fields were involved the patients appeared to be more ill.

Etiology.—No specific organisms could be isolated from those patients on whom bacteriologic studies were attempted. Viral causation was suggested by the absence of leukocytic response to the infection and by the failure of sulfonamide therapy. Recent reports have strengthened the case for a theory of viral causation. Viruses have been isolated from normal mouse lungs which required only serial passage through other mouse lungs to become pathogenic.[10] This result suggests the possibility of a human reservoir of the virus among healthy persons. Serums of some patients in New York city gave positive neutralization reactions against psittacosis virus.[11] Not only have viruses been isolated from some patients,[12] but the Commission on Acute Respiratory Diseases[13] has been able to transmit the disease from throat washings and sputum of patients to volunteers. Since both untreated and filtered material were equally successful, a filtrable agent would appear to be responsible.

Differential Diagnosis.—The lack of pathognomonic history or signs makes differential diagnosis difficult, especially in the early stages of the illness. The only diagnostic laboratory observation, the demonstration of an increased titer of cold agglutinins in the serum, is of limited value. They are frequently absent, as in my patients, and even when they occur their presence cannot be detected until after the acute phase of the disease has passed. These difficulties further emphasize the importance of the roentgenogram of the chest in establishing the nature of the illness. The disease closely resembles influenza in its onset and early course; in the presence of cough and signs referable to the chest, acute bronchitis is suggested. It is in several respects distinctly unlike influenzal pneumonia as seen during the last war,[14] the latter having been clinically more severe, often with a characteristic cyanosis, an elevated white blood cell count associated with secondary bacterial infection and a high mortality.

The severe frontal headache, chills and fever, generalized aching and unelevated white blood cell count frequently make it necessary, during the warmer months in Italy, to rule out malaria and sandfly fever.

8. Duggan and Powers.[4b] Whiteley, Bernstein and Goldman.[4e] Green and Eldridge.[4g] Dingle and others.[1] Young, Storey and Redmond.[4c]

9. Bahlke, A. M.: New York State J. Med. **43**: 315 (Feb. 15) 1943.

10. Horsfall, F. L., Jr., and Hahn, R. G.: J. Exper. Med. **71**:391 (March) 1940.

11. Smadel, J. E.: J. Clin. Investigation **22**:57, 1943.

12. (*a*) Reimann, H. A.: Acute Infection of Respiratory Tract with Atypical Pneumonia: Disease Entity Probably Caused by Filtrable Virus, J. A. M. A. **111**: 2377 (Dec. 24) 1938. (*b*) Stickney, J. M., and Heilman, F. R.: Proc. Staff Meet., Mayo Clin. **17**:369 (June 17) 1942. (*c*) Beck, M. D., and Eaton, M. D.: J. Infect. Dis. **71**:97 (Sept.) 1942.

13. Transmission of Primary Atypical Pneumonia to Human Volunteers: Commission on Acute Respiratory Diseases, J. A. M. A. **127**:146 (Jan. 20) 1945.

14. Reimann, H. A.: Bull. New York Acad. Med. **19**:177 (March) 1943.

Most important from the viewpoint of therapy is the differentiation from pneumococcic and other bacterial pneumonias. Lobar pneumonia can usually be recognized as such in its sudden onset and marked pulmonary symptoms and signs; certain bronchopneumonias, especially those with a rapid infiltrative spread from the upper respiratory passages, are much more difficult to differentiate. In primary atypical pneumonia, the mode of onset, repeated chilly sensations, dry cough and frequent pain in the chest in the absence of distinct physical signs, combined with a normal white blood cell count, are characteristic. The roentgenogram is most important but may not show changes early in the course of the disease.[15] When doubt exists as to the nature of the pneumonia, the patient should be given the benefit of it in the form of a chemotherapeutic test. Though prevailing opinion considers the sulfonamide compounds of no value, there are some [5c] who urge their use to forestall secondary bacterial invasion. In any event, there is in the disease itself no contraindication to chemotherapy. If no response is obtained after two to three days of full doses and of adequate blood levels of the drug, it may be discontinued.

Therapy.—I have not been aware of any therapy which considerably influences the course of the disease. Sulfonamide compounds have repeatedly been without effect. More recently, penicillin proved to be equally ineffectual. Plasma and whole blood were given to some of the more seriously ill patients, without noticeable improvement. Symptomatic treatment during the acute phase consisted of rest, high fluid intake (parenterally when necessary), alcohol sponges, codeine and acetylsalicylic acid for head and body aching and expectorants. When pleural pain was severe, procaine hydrochloride was injected locally at the proper level. Inhalation of oxygen was used in but a few cases for difficulty in breathing. During the slow convalescence, supportive therapy in the form of a high caloric diet supplemented by multivitamin capsules was given. Activity was increased gradually without necessarily waiting for complete roentgenologic resolution.[16] After being allowed to be out of bed for some days, patients were permitted to go to mess and then to morning exercises for several days prior to discharge. I was not in a position to use roentgen ray therapy as recommended by Oppenheimer [17] and Hufford and Applebaum.[18] Readiness for discharge was based on the roentgenogram of the chest, the erythrocyte sedimentation rate and the general appearance, particularly as reflected in the gain in weight, of the patient. Hospitalization was usually of four to seven weeks' duration.

Complications.—There were no fatalities in my series. Complications were uncommon. Eight patients were left with a thickened interlobar pleura and 1 with diaphragmatic pleural adhesions. One patient had a moderately severe earache without apparent cause, which subsided spontaneously. Two patients had herpes labialis; 2 had associated malaria. I did not encounter any thrombophlebitis or chronic productive cough.

REVIEW OF THE LITERATURE

A review of a number of reports on primary atypical pneumonia reveals, as indicated by Reimann, Havens and Price,[19] that the disease—if one is to regard all descriptions as a single disease entity—occurs in two principal forms: (1) a sporadic, more severe illness encountered mostly in civilian practice [20] and (2) an epidemic, milder form encountered in institutional life in schools, army camps and hospitals, usually observed, therefore, among younger persons. This form is described by most authors.[21] It would seem that younger persons are better able to combat the disease and to avoid long sieges of illness and complications. Even in this young age group, the slowest recoveries were seen in a few of the older patients. The cases were of the milder type of the disease, and, though they resembled other series from army sources, they differed in a few respects.

15. (*a*) Smiley, D. F.; Showacre, E. C.; Lee, W. F., and Ferris, H. W.: Acute Interstitial Pneumonitis: New Disease Entity, J. A. M. A. **112**:1901 (May 13) 1939. (*b*) McCarthy, P. V.: Radiology **40**:344 (April) 1943. Thompson.[5c]

16. (*a*) Drew, W. R. M.; Samuel, E., and Ball, M.: Lancet **1**:761 (June 19) 1943. (*b*) van Ravenswaay, A. C., and others: Clinical Aspects of Primary Atypical Pneumonia: Study Based on 1,862 Cases Seen at Station Hospital, Jefferson Barracks, Missouri, from June 1, 1942 to Aug. 10, 1943, J. A. M. A. **124**:1 (Jan. 1) 1944.

17. Oppenheimer, A.: Am. J. Roentgenol. **49**:635 (May) 1943.

18. Hufford, C. E., and Applebaum, A. A.: Radiology **40**:351 (April) 1943.

19. Reimann, H. A.; Havens, W. P., and Price, A. H.: Etiology of Atypical ("Virus") Pneumonias, with Brief Résumé of Recent Discoveries, Arch. Int. Med. **70**:513 (Oct.) 1942.

20. Reimann.[12] Kneeland and Smetana.[4f] Longcope.[8a]

21. (*a*) Goodrich, B. E., and Bradford, H. A.: Am. J. M. Sc. **204**:163 (Aug.) 1942. (*b*) Haight, W. H., and Trolenger, J. H.: U. S. Nav. M. Bull. **41**:988 (July) 1943. (*c*) Smiley and others.[15] Maxfield.[2a] Duggan and Powers.[4b] Dingle and Finland.[3b] Whiteley, Bernstein and Goldman.[4e] Green and Eldridge.[4g] Dingle and others.[1] Correll and Cowan.[5d] Young, Storey and Redmond.[4c] van Ravenswaay and others.[16b]

It is probable that many cases diagnosed as influenza during World War I were actually cases of atypical pneumonia. At Fort Sam Houston, for example, one fourth of 1,000 patients with "uncomplicated influenza" had persistent basal rales, and roentgenographic examination revealed local areas of increased density in almost all this group.[22] Bowen,[23] in 1935, described the characteristic roentgenographic picture in what appears to have been a mild form of the illness, and the following year Allen,[24] again from Fort Sam Houston, reported 68 cases of "acute pneumonitis" with similar roentgenographic features. Clinically, the cases were characterized by a "benign course, few physical signs, and roentgenologic evidence of localized inflammatory process in the lung."

Primary atypical pneumonia, as described by the Commission on Pneumonia for the Surgeon General,[4d] bears many similarities to the illness in my patients as well as some points of difference. The described chilly sensations, dry cough later becoming productive, relative bradycardia, normal or slightly increased respiratory rate, involvement of the lower lobes with a few physical signs relative to the roentgenographic signs, normal white blood cell count, duration of fever and prolonged convalescence disproportionate to the apparent severity of the disease were all evident in this series. However, the gradual onset with only mild headache and absence of pleuritic pain differ from the symptoms of my patients. The cases of Moore, Tannenbaum and Smaha[25] were similar to some of mine in which the patients were less seriously ill, having febrile periods of two to five days with complete resolution in three or four weeks. In the 80 cases reported by Green and Eldridge,[4g] as in my own, the patients exhibited intense frontal headache, cough, pain in the chest and meager signs referable to the chest; one third of the patients had a mild tachycardia during convalescence. Though pain in the chest was present in 35 per cent of their patients, pleuritic pain was described as rare. Another series of cases of military origin, that of Whiteley, Bernstein and Goldman[4e] resembled mine in the lack of definite physical signs, the production of blood-flecked sputum and fever of seven to ten days' duration. In these too, unlike others, pleuritic pain was rare; a less common feature here reported was a definite leukopenia in all but 1 case.

Duggan and Powers,[4b] from their experience with naval personnel, described an illness resembling that which I observed in its griplike onset, minimal physical signs with coarser rales after defervescence, relative bradycardia, roentgenographic changes, normal blood count and absence of complications. Their patients were apparently less severely ill than mine, requiring less hospitalization. No pleuritic pain or hemoptysis was encountered, and in three fourths of the patients, the chest was normal on physical examination. Correll and Cowan,[5d] reporting 155 cases, described an abrupt onset with minimal physical signs in all patients and predominance of pathologic conditions in the lower lobes. Another report from naval sources by Haight and Trolenger[21b] described cases similar to mine.

In the study of Dingle and his associates[1] three fourths of the patients had a gradual onset of the illness and 35 per cent a preceding infection of the upper respiratory tract. Chilliness was experienced by 75 per cent, but only 13 per cent had shaking chills. Cough was present in all patients and blood-streaked sputum in 12 per cent. Pain in the chest was present in 44 per cent; in 18 per cent it was pleuritic in nature. Positive signs referable to the chest, chiefly sticky, subcrepitant rales at the end of inspiration, were heard in less than half of the patients. The roentgenograms, similar to those of my patients, showed involvement of the lower lobe in 70 per cent. Stressed also was the lack of correlation betwen physical signs and extent of roentgenographic changes. The signs often appeared late, when the temperature had begun to subside. The white blood cell count remained normal as a rule. Duration of hospitalization averaged 31.7 days (compared with 36 days in my series). The only complication of any consequence was a chronic productive cough, suggestive of early bronchiectasis but not proved. Other complications reported from time to time, such as thrombophlebitis, pleural effusion and manifestations referable to the central nervous system, were not encountered. During the period of study, 34 cases of "bronchitis resembling atypical pneumonia," but lacking roentgenographic corroboration, were observed; these were considered to represent a mild form of the disease.

One of the most comprehensive reports is that of van Ravenswaay and others,[16b] based on 1,862 cases. In this group, the onset was sudden in one third and gradual in two thirds. Chilliness was experienced by 68 per cent but associated

22. Hall, M. W.: Inflammatory Diseases of the Respiratory Tract, in Ireland, M. W.: The Medical Department of the United States Army in the World War, Washington, D. C., Government Printing Office, 1928, vol. 9, p. 152.

23. Bowen, A.: Am. J. Roentgenol. **34**:168 (Aug.) 1935.

24. Allen, W. H.: Ann. Int. Med. **10**:441 (Oct.) 1936.

25. Moore, G. B., Jr.; Tannenbaum, A. J., and Smaha, T. G.: Atypical Pneumonia in Army Camp, War Med. **2**:615 (July) 1942.

rigor by only 11 per cent. Pain in the chest was present in 70 per cent and seemed of pleural origin in 48 per cent. The value of the erythrocyte sedimentation rate as a means of evaluating the course of the disease was stressed—an observation with which I fully agree. A pertinent point concerning the length of hospitalization was made. Though complications occurred largely in those patients who were allowed to be ambulatory soon (four afebrile days), it was not necessary to wait until the roentgenogram showed complete clearing. Patients with two weeks of normal temperature and a low erythrocyte sedimentation rate could tolerate exercise without reactivation or impaired clearing of the roentgenographic picture.

An outbreak of 40 cases of the disease among hospital personnel described by Young, Storey and Redmond [4c] showed many features similar to the ones described here. Chilly sensations, persistent cough, headache and generalized aching were common. These authors stressed also the relative bradycardia with increase in the rate at lysis, an increase in respiratory rate only in extensive involvement, fever of five to eleven days, usual involvement of the lower lobes and a lack of complications. Another localized appearance of the disease, 40 cases representing 40 per cent of a company, was described by Idstrom and Rosenberg.[2] All cases appeared in ten days. As in this series, the picture included severe frontal headache, dry cough, seven to ten days of fever, few physical signs until the fifth or sixth day, a sputum negative for pathogenic organisms, absence of cold agglutinins and an elevated erythrocyte sedimentation rate whose fall correlated well with the clearance of the chest. Hospitalization lasted thirty to forty days, and there was no fatality.

The disease as described by Reimann,[12a] Kneeland and Smetana [4f] and Longcope [5a] appeared to be of the more severe variety with greater toxicity, longer duration, more complications and higher fatality. Longcope, in fact, divided his cases into three groups according to the severity of the disease, namely, mild, moderately severe and severe. The first group was the largest, included half his cases and most approximated the disease as seen in my patients. These severe forms of the disease occurred for the most part in sporadic cases among older persons in the civilian population.

CONCLUSIONS

Primary atypical pneumonia is an acute infectious, usually self-limited disease occurring in varying severity. In the milder form, there are two distinct phases of the disease: namely, a short acute febrile period lasting seven to ten days and a prolonged interval of convalescence during which the pathologic process slowly recedes and the absorption of pathologic products coincides with an increase in heart rate and white blood cell count and a prolonged elevated erythrocyte sedimentation rate. Though the disease itself is old and has probably been identified in the preserved lungs of American Civil War soldiers, the growth of our understanding of the disease during the past five years stems historically from two facts.[26] The increased use of the roentgenogram has demonstrated a picture that is characteristic and differing in its development, course and resolution from that of bacterial pneumonia and, in some cases, has revealed the presence of the disease in the absence of other evidence. Also, the use of sulfonamide compounds has been followed by a condition leading to a better understanding of the disease. A larger number of patients with pneumonia were treated in the home; those patients who failed to respond to treatment were hospitalized. In short, relatively more patients with primary atypical pneumonia reached those institutions in which the disease could be best studied and segregated.[14] The high incidence of the disease in the services also added to the knowledge concerning it.

The nature of the pathologic process has, in several ways, made the disease difficult to diagnose. The lesions' being interstitial rather than exudative into the air spaces results in the absence of dyspnea, tachypnea and cyanosis and minimizes physical signs, thereby focusing attention away from the chest. Moreover, even though the clinician be well aware of the early symptoms and be prompted to order that a roentgenogram be made, he is apt to be misled by the delay in appearance of roentgenologic changes unless he repeats the study after several days. The tendency on viewing the first, normal roentgenogram is to regard the condition as an infection of the upper respiratory tract and to discharge the patient shortly after the fever has subsided, just when continued rest is of great importance in preventing the spread of the pneumonic process. A greater awareness of this condition is still necessary.

Proper treatment of the disease requires an understanding not only of the nature and high incidence of the illness but also of its pathology and of the lack of correlation between physical and roentgenologic signs and of the course of the disease process.

Lieutenant Colonel W. W. Bondurant Jr., Army of the United States, cooperated in this study.

26. Spink, W. W.: Minnesota Med. **26**:337 (April) 1943.

ACUTE PORPHYRIA

CLINICAL AND PATHOLOGIC OBSERVATIONS

CAPTAIN MAX BERG*

MEDICAL CORPS, ARMY OF THE UNITED STATES

With the increasing knowledge of the importance of porphyrin metabolism and with the increasing frequency with which porphyria has been reported, there has been widespread interest in this fundamental subject. There is lack of knowledge, however, of the biologic mechanisms involved as well as of the cause of the varied symptoms. In this connection I thought that the results of my observations, both clinical and pathologic, might be of interest.

The porphyrins are widely distributed in nature.[1] They form part of the complex molecule of hemoglobin, myoglobin and cytochrome. The porphyrins arise in the body during synthesis of hemoglobin rather than during its destruction, as had been previously assumed.[2] A number of excellent comprehensive reviews of the subject of porphyrins have appeared in the recent literature,[3] some of the reports being particularly pertinent to this paper.

Small amounts of porphyrin of the isomeric series I and II are excreted as coproporphyrin I and III in the urine of normal persons. While porphyrin I is not utilized and is excreted in the urine as coproporphyrin I, the porphyrin III which is not utilized, owing to its being synthesized in amounts greater than that required to enter into the formation of hemoglobin or owing to a possible toxic block,[4] is excreted as coproporphyrin III. In porphyria as well as in a great variety of other diseases, these pigments are excreted in amounts far above the normal range.

The forms of porphyria have been classified into three broad groups: congenital, acute and chronic. While these represent a general classification, some striking variations in the symptoms may be observed, and occasionally the case may present features of more than one group. This variability was illustrated in the case to be described, which presented some of the clinical features found in congenital as well as in acute porphyria.

In congenital porphyria, the onset is usually early in life but may be delayed until the patient is 15 years of age. There is a familial tendency. Photosensitivity is prominent, and a chronic bullous or erythematous dermatitis with exacerbations in the spring and summer is present. The urine is burgundy red at times, with large amounts of coproporphyrin I or uroporphyrin I or III. At autopsy, very little may be observed except a pinkish discoloration of the teeth and bones.

In acute porphyria the attacks are frequently characterized by a sudden onset, with severe abdominal pain and obstipation. The symptoms[3b] may be classified into three types: (1) abdominal, (2) nervous and (3) mental. They may occur singly or may be combined. Frequently the acute attacks are ushered in with the menstrual period, or they may occur post partum. It has recently been pointed out[5] that acute porphyria is as much a familial disease as congenital porphyria, with the underlying mechanism an inborn error of pigment metabolism.

Hypertension and tachycardia are frequent. There is often a low grade fever, and leukocytosis may be present. Nervous symptoms, such as severe pains and weakness in the extremities, bulbar weakness and a symmetric paralysis of the lower motor neuron type with flaccidity and loss of reflexes, may develop.[6] A classic Landry

* Formerly from the Department of Pathology, University of Illinois College of Medicine, Chicago.

1. Dobriner, K., and Rhoads, C. P.: The Porphyrins in Health and Disease, Physiol. Rev. **20**:416-468 (July) 1940.

2. Nesbitt, S.: Excretion of Coproporphyrin in Hepatic Disease, Arch. Int. Med. **71**:483-488 (April) 1943.

3. (*a*) Dobriner and Rhoads.[1] (*b*) Waldenström, J.: Studien über Porphyria, Acta med. Scandinav., 1937, supp. 82, pp. 1-254. (*c*) Watson, C. J.: Porphyrins and Diseases of the Blood, in Symposium on Blood, Madison, Wis., University of Wisconsin Press, 1939, pp. 14-30. (*d*) Mason, V. R.; Courville, C. B., and Ziskind, E.: The Porphyrins in Human Disease, Medicine **12**:355-439 (Dec.) 1933.

4. Dobriner and Rhoads.[1] Watson.[3c] Nesbitt.[2]

5. Nesbitt, S.: Acute Porphyria, J. A. M. A. **124**: 286-294 (Jan. 29) 1944.

6. Hoagland, P. I.: Acute Porphyria: Report of Two Cases with Neurologic Manifestations, Proc. Staff Meet., Mayo Clin. **17**:273-281 (May 6) 1942.

type of paralysis has been described. With the development of neurologic manifestations, a mortality as high as 75 per cent may be expected. Remissions and improvement do occur, particularly with the abdominal type, in which recurrences are frequent. Occasionally various mental changes [6] are observed. The urine may be burgundy red, or it may be amber, but on exposure to sunlight it changes to the characteristic color. A simple test for porphyrins in the urine has been recently described by Watson and Schwartz.[7] The increased porphyrins in the blood may be detected by a number of methods. Coproporphyrin I and III have been observed in the feces.[8] The pathologic observations are meager, but the most extensive descriptions have been presented by Waldenström [3b] and Mason, Courville and Ziskind.[3d]

Acute porphyria has been further classified into acute toxic and acute ideopathic forms; however, these forms apparently present no fundamental clinical differences,[3] the differentiation being made on the history of an exciting etiologic agent. Chronic porphyria is not well characterized and possibly does not represent a distinct form but may constitute a mild form of either congenital or acute porphyria.

It was thought of interest to study the reaction of the capillaries, gastrointestinal motility, and blood chemistry and to attempt to correlate these factors with some of the protean symptoms of porphyria. Rusk and Howell [8] showed that in experimental animals injections of hematoporphyrin had a pronounced effect on the circulation, particularly the cutaneous circulation, with a gradual rise in blood pressure beyond the normal followed by a fall, vascular collapse and death. There was presented evidence that the hematoporphyrin combined slowly with a substance or substances in tissues and that this compound was formed in the dark as well as in the light. It was the action of light on this combination that gave rise to the injurious effects. The action of the porphyrins on the gastrointestinal tract has been variously ascribed by different authors to different mechanisms; that is, the disturbed function has been thought to be due to lesions of the sympathetic ganglions (Mason, Courville and Ziskind [3d]) or to a toxic effect on muscles which is not abolished by atropine (Dobriner and Rhodes [1]) or to the action on the autonomic nervous system.[5]

7. Watson, C. J., and Schwartz, S.: A Simple Test for Urinary Porphyrobilinogen, Proc. Soc. Exper. Biol. & Med. **47**:393-394 (June) 1941.

8. Rask, E. N., and Howell, W. H.: Photodynamic Action of Hematoporphyrin, Am. J. Physiol. **84**:363-377 (March) 1928.

REPORT OF A CASE

The patient, a 20 year old woman, was referred to me by Dr. M. Waisman because of the sudden development of severe abdominal pains, nausea and persistent vomiting.

She gave a history of being entirely well until the age of 16, when, after the onset of menstruation, she noted the occurrence of a bullous eruption over the exposed portions of the body. The condition was worse in the spring and summer, when she blistered easily on exposure to the sun. The cutaneous condition was later diagnosed as epidermolysis bullosa.

In March 1940 she had her first attack of generalized abdominal pain and persistent vomiting. An exploratory operation was performed, and a normal appendix was removed. After the operation the complaints gradually subsided.

In October 1940 she had another attack of abdominal pain and persistent vomiting, and jaundice developed. This was associated with a mild psychotic episode. This acute phase persisted for a period of seven weeks, and the diagnosis of acute porphyria was established.[9] The urine was reddish brown and contained large amounts of coproporphyrin and uroporphyrin as the zinc metal complex. A decided increase of porphyrins was also demonstrated in the blood. The bilirubin of the blood increased to 19.8 mg., and the van den Bergh reaction was direct. The serum protein was 8 Gm. per hundred cubic centimeters, and the albumin-globulin ratio was 1.1 to 1. Tests of sensitivity to light showed the patient to be sensitive to the far ultraviolet portion of the spectrum.

The urinary excretion of porphyrin varied considerably from time to time, and a urinary excretion of coproporphyrin up to 3 mg. in twenty-four hours was reported [10] early in 1941.

When first seen, on May 29, 1941, in addition to having pains and vomiting of two days' duration, the patient felt cold in spite of the intense heat (the temperature ranging in the nineties), and she complained of severe pains in the knee joints. On physical examination the pulse rate was found to be 120, the temperature 99 F., and the blood pressure 130 systolic and 110 diastolic. The abdomen was soft; there was no rigidity or tenderness, and the peristaltic sounds were diminished. The remainder of the physical and neurologic examination showed nothing abnormal. As there is no specific treatment for acute porphyria besides general supportive measures, various attempts were made to control the course of the symptoms. The first day, the patient was given 10 cc. of a 10 per cent solution of calcium gluconate by injection with no demonstrable effect, and morphine was given to control the pain.

The variations in blood pressure and pulse rate may be observed in figure 1. The next day, May 30, her condition remained unchanged. She was given 1 cc. of a solution of ergotamine tartrate hypodermically, after which she felt warm and began to perspire; the abdominal pain persisted, however. The following day the vomiting diminished; otherwise the condition remained unchanged. She was then given atropine, 0.45 mg. orally four times daily, with complete subsidence of the vomiting; however, the pains persisted. On

9. Nesbitt, S., and Watkins, C. H.: Acute Porphyria, Am. J. M. Sc. **203**:74-83 (Jan.) 1942. Nesbitt, S.: Personal communication to the author.

10. Cornbleet, T.: Personal communication to the author.

June 1, trasentin (diphenylacetyl-diethylaminoethanol hydrochloride) tablets were substituted for the atropine. On the following day it was noted that the pains were worse and that the pulse rate and diastolic blood pressure were still elevated, with the pulse rate being 130 and the blood pressure 120 systolic and 96 diastolic. She was given 1 cc. of a 1 : 2,000 solution of neostigmine bromide subcutaneously, and in one hour there was a definite diminution of the abdominal pains, which continued to subside. The next day, on examination, she showed a pronounced improvement, the pulse rate and blood pressure having dropped to normal. She complained of only an occasional pain. Another cubic centimeter of neostigmine solution (1 : 2,000) was administered, and the patient felt entirely well.

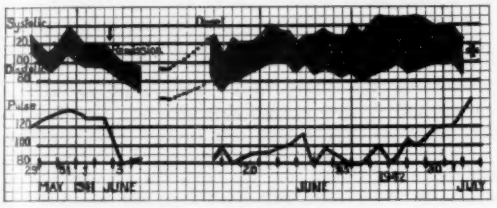


Fig. 1.—Daily record of the blood pressure and pulse rate.

The significant laboratory observations were: a pinkish color of the urine, which turned darker on standing; erythrocyte count, 4,200,000; hemoglobin content, 85 per cent; leukocytes, 5,500; neutrophils, 63 per cent, and lymphocytes, 36 per cent.

In the interval succeeding this attack, the patient was seen on two occasions. Except for her cutaneous lesions, she felt entirely well, and the examination showed nothing abnormal.

The observed acute attack was characterized by persistent vomiting, abdominal pain and diminished peristaltic sounds on auscultation. Further, there were present vasomotor phenomena, such as an increased blood pressure, particularly the diastolic, the sensation of severe cold in spite of the intense heat, lack of perspiration and articular pains. The question raised was whether the improvement which was observed after the injection of neostigmine bromide was a chance coincidence, for spontaneous remissions of the acute attacks do occur, or whether there was a definite causal relation, for the remissions do not occur as abruptly as did the one observed. In order to answer this question, it was decided to postpone reporting these observations until they could be confirmed either on other patients or on this patient during another attack. In addition, it was thought of interest to study reactions of the capillaries, gastrointestinal motility and blood chemistry and to attempt to correlate these with the symptoms. In another acute attack involving the same patient the following observations were made.

On June 17, 1942, the patient again had an acute attack similar to the one previously observed. The onset occurred with the menstrual period, as in the previous attack. The external temperature again was high, although she complained of being chilled. On physical examination the pulse rate was found to be 90 and the blood pressure 130 systolic and 90 diastolic. The skin was dry and the abdomen soft, and peristaltic sounds were absent. The patient was given 1 cc. of a solution of neostigmine (1 : 2,000) hypodermically, after which she began to perspire; there was some diminution of abdominal pain, and peristaltic sounds were heard over the abdomen. The articular pains were unaffected. On June 19 the condition was the same as that originally observed. She was given 1 cc. of the neostigmine solution and 6 mg. of mecholyl bromide, which produced severe vomiting, salivation, a feeling of heat replacing the feeling of cold, profuse perspiration and complete relief from the abdominal and the articular pains. There was a drop in blood pressure and an increase in pulse rate. After three hours the articular pains recurred.

On June 20 it was noted that after the injection the vomiting ceased, and it never recurred; considerable peristalsis was present over the abdomen. The patient was given 3 tablets of neostigmine bromide (15 mg. each) daily and 1 cc. of the solution by injection. The following day the abdominal pains recurred, with diminished peristalsis. After injection of 1 cc. of neostigmine solution there was a diminution of pain and increased peristalsis.

For the next six days she continued to improve symptomatically. By June 28 the abdominal pains had subsided completely, and the patient felt well; however, the blood pressure was persistently elevated, being 148 systolic and 100 diastolic, and the pulse rate was rapid. There was an icteric tinge to the scleras. The next day she complained of some weakness in the arms and legs, and in the evening she spoke irrationally. The following day, June 30, a definite increase in muscular weakness was present. The weakness in the arms and legs was more pronounced in the proximal than in the distal segments. She could move only her toes, fingers and hands. Tendon reflexes and the Babinski sign were absent. Sensibility to pain was intact as well as the position sense in fingers and toes. She was given 1 cc. of neostigmine solution and 1 mg. of mecholyl, with a transient increase in muscular strength.

The patient became rapidly weaker; the pulse rate increased. Finally she showed difficulty in phonation; Cheyne-Stokes respiration developed, and she died on July 2, 1942.

Laboratory Observations.—The observations are summarized in the table. In addition, the blood exhibited a red fluorescence indicative of the presence of porphyrin.[11] The value for serum protein was elevated, as was evidenced in a previous attack.[11]

The albumin-globulin ratio of 1.1 to 1 was also similar to that observed in a previous attack. After the

Fig. 2.—Kymographic record of gastric contractions. Record to the left shows gastric atony; after *P* (injection of 1 cc. of 1 : 2,000 solution of neostigmine bromide) gastric contractions are seen. The white dots below represent time in minutes.

administration of mecholyl and neostigmine, the only significant change in the blood chemistry was an increase in the blood sugar.

An interesting observation was the reduction in the coagulation time (measured by Howell's method). Al-

11. Mason, H. L.: Personal communication to the author.

though there was no evidence of thrombosis at autopsy, difficulty was experienced in inserting the needle into the peripheral veins while the patient was under observation.

Gastric Motor Activity.—At the onset of the last attack, before any medication was administered, a balloon was passed into the stomach in order that a recording of gastric contractions might be made. Varying reports of increased or diminished activity as observed fluoroscopically may be found in the literature.[3] The kymographic tracing may be observed in figure 2. During a sixty minute control period, the stomach remained dilated, and no gastric contractions were recorded. After the injection of 1 cc. of neostigmine solution (1:2,000) in one and one-half minutes, gastric contractions were recorded, which increased in amplitude to reach a maximum. At first the waves were regular, and then they became irregular, although the height of the contractions remained the same.

Observations on the Capillaries of the Skin.—The capillaries were observed through a capillary microscope. At the onset, in observations on the hand under low magnification, the skin appeared pale, and the capillaries appeared either as faint violaceous loops or as dots. From one to three capillaries were visible per low power field. On the nail bed four capillaries were visible per low power field. They were short and extremely dilated and presented the picture of stasis. In only one of the four capillaries in a field was movement of blood seen. In some of the capillaries an aneurysmal dilatation was present. Within a short time after the injection of solution of neostigmine, very little change in the capillaries was seen. A number of hours later considerable capillary activity was observed, and again a few showed an aneurysmal dilatation. Although the capillaries were slightly increased in number, they were still much fewer than the number observed in the normal skin.

After the injection of both neostigmine and mecholyl solution, a pronounced increase in the number of capillaries was visible; the number increased to fifteen to twenty per low power field. They were of various shapes and showed considerable activity. On the nail bed many capillaries were seen; they were tortuous and active. Even though they were dilated, red blood cells could be observed moving rapidly along.

Results of Determinations on the Blood

	Blood p_H	Serum Protein (Refractometer)	Serum Globulin	Serum Albumin	Dextrose	Calcium	Non-protein Nitrogen	Coagulation Time (Howell's Method)	White Blood Cells	Red Blood Cells
June 18, 1942.....	7.43	9.49	3.60	4.00	80	....	28	40 sec.	.....	.....
	7.42	8.96	....	....	89	8.69	..	35 sec.	5,050	4,390,000
After neostigmine	7.42	8.68	....	....	95	....	..	2 min.	5,300	3,310,000
June 19, 1942.....	7.34	9.50	....	....	109	....	..	1 min. 50 sec.	4,000	4,220,000
After neostigmine and mecholyl.....	7.46	9.90	....	....	138	....	..	35 sec.	6,500	4,600,000

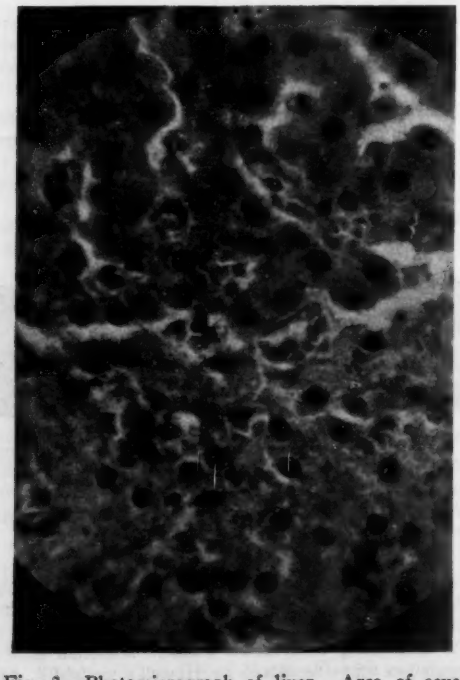


Fig. 3.—Photomicrograph of liver. Area of severe degenerative changes, sharply demarcated, is seen in the lower half of the field. (× 400)

Autopsy Observations.—Gross observations revealed very little except a pinkish tinge of the tissues. The cartilage and the tissues of the breast were pink, and the bone presented reddish discolorations. The brain and spinal cord were reported by Dr. Percival Bailey as being essentially normal. Microscopically, the liver showed diffuse degenerative changes, extreme granular cytoplasmic swelling and some vacuolation, with small localized areas of normal hepatic tissue. The hepatic cells in both the normal and the degenerated areas showed a yellowish pigment, with the amount of pigment being much less in the normal areas. The Kupffer cells were swollen and filled with a non-iron-containing pigment. Some of the hepatic changes may be observed in the photomicrograph (fig. 3).

In the kidneys a number of the glomeruli showed a definite thickening of Bowman's capsule; otherwise, there were no significant changes.

Except for focal areas of bronchiolar pneumonia in the lungs, the other organs were normal.

COMMENT

The primary disturbance was a decided increase of the porphyrin in the blood. Coproporphyrin was excreted by the liver and kidneys, the liver being the more important organ of excretion.[12] Severe parenchymatous damage to

12. Nesbitt.[2] Nesbitt, S., and Snell, A. M.: The Excretion of Coproporphyrin in Hepatic Disease: I

the liver was observed, with focal areas of relatively normal hepatic tissue remaining. With increasing parenchymatous hepatic damage, massive amounts of coproporphyrin were excreted in the urine by a mechanism similar to the excretion of bile pigments.[12]

The pressor episode, that is, the episode characterized by elevated blood pressure, sensation of cold and increased pulse rate, was probably a result of the extreme capillary spasm observed in the skin. Injury to the capillaries in the form of aneurysmal dilatations was observed. While no similar observations have been reported in the literature, Waldenström[3b] has reported changes in the arterioles in the form of subacute inflammatory changes, medial necrosis and sclerosis in several cases of porphyria. These aneurysms in the capillaries of the skin may be the result of the long-continued action of the porphyrins or the result of the interaction of sunlight on the tissues by a mechanism similar to that described by Rask and Howell.[8] The bullae arising in the skin as a result of exposure either to sunlight or to direct trauma may be related to the increased permeability of the capillaries, which in turn is the result of the injury to the capillary. Further studies are necessary to answer this question. The pains in the joints appear to have been due to capillary spasm with possible microscopic hemorrhages, for with release of the spasm immediate relief of the pain was observed.

At the same time that the capillary spasm was observed, a noticeable disturbance in gastric contractility was seen in the form of a prolonged period of atony, interrupted only by a brief rush of reverse peristalsis. In addition, there was an absence of peristaltic sounds over the abdomen. Immediately after the injection of neostigmine solution, the resumption of gastric contractions was recorded. The action on the capillaries was less noticeable. With the combined injection of mecholyl and neostigmine solutions, a pronounced reaction occurred, with definite capillary dilatations and gastric and intestinal contractions. Associated with these there was complete symptomatic relief of varying duration. As the reported action of neostigmine is to inhibit the destructive action of cholinesterase on acetylcholine and the action of acetylcholine (to which mecholyl is closely related) is manifested by parasympathetic stimulation and is necessary for the transmission of nervous impulses,[13] the question arises as to the action and the site of action of the porphyrins as well as to their relation to the action of the drugs just mentioned.

While considerable research must be done to explain the action of the porphyrins on the nervous system, there is evidence to suggest several hypotheses. The porphyrin may produce a block in neuromuscular transmission and inhibit the action of acetylcholine at synapses, parasympathetic nerve endings and myoneural junctions and in the blood. In this connection, while the porphyrins circulate in the blood, the assumption of specificity and localization of action must be made. In support of this hypothesis may be cited by analogy the report of Kabat and Knapp,[14] who observed many examples of localization and specificity of the action of neostigmine in patients with poliomyelitis, although the drug is presumably distributed evenly throughout the body. Further support of the concept of a toxic block might be assumed in the results of recent observations in myasthenia gravis,[15] concerning which it is believed that the muscular weakness is due to some substance in the blood, possibly secreted by the thymus gland, which produces a block in neuromuscular transmission and inhibits the normal action of acetylcholine on voluntary muscle. The action of neostigmine in myasthenia gravis, according to this assumption, has been thought to produce some antagonistic effect on this inhibiting substance and to restore the normal release and rate of hydrolysis of acetylcholine. A possible action of the porphyrins may be to interfere with the control of the rate of hydrolysis of acetylcholine and with its relation to cholinesterase. Another possibility is that the porphyrins may interfere with the mechanism of synthesis and release of acetylcholine as postu-

Urinary and Fecal Excretion as Correlated with Parenchymatous Hepatic Damage, Arch. Int. Med. **69**:573-581 (April) 1942; II. Urinary and Fecal Excretion in Biliary Obstruction, ibid. **69**:582-588 (April) 1942.

13. Sollmann, T.: A Manual of Pharmacology, ed. 6, Philadelphia, W. B. Saunders Company, 1942. Lorente de Nó, R.: Liberation of Acetylcholine by the Superior Cervical Ganglion and the Nodosum Ganglion of the Vagus, Am. J. Physiol. **121**:331-349 (Feb.) 1938. Nachmansohn, D., and Meyerhof, B.: Relation Between Electrical Changes During Nerve Activity and the Concentration of Choline Esterase, J. Neurophysiol. **4**:348-361 (July) 1941.

14. Kabat, H., and Knapp, M. D.: The Use of Prostigmine in the Treatment of Poliomyelitis, J. A. M. A. **122**:989-995 (Aug. 7) 1943.

15. Wilson, A., and Stoner, H. B.: Myasthenia Gravis: A Consideration of Its Causation in a Study of Fourteen Cases, Quart. J. Med. **13**:1-18 (Jan.) 1944. Harvey, A. M.; Lilienthal, J. L., Jr., and Talbot, S. A.: Observations on the Nature of Myasthenia Gravis: The Effect of Thymectomy on Neuromuscular Transmission, J. Clin. Investigation **21**:579-588 (Sept.) 1942.

lated in recent reports.[16] A third possibility is that the action of the porphyrins on the nervous system may be the result of a combination of these factors, that is, a possible interference with the mechanism of the formation and release of acetylcholine [16] and an interference with the control of the rate of the hydrolysis of acetylcholine and with its relation to cholinesterase.[13] Variations must be assumed in the severity of action as well as in the site of action, whether the involvement be the peripheral or the central neurons, the preganglionic or the postganglionic nerves, the nerve synapses or the neuromuscular plates; for various reports have told of autonomic dysfunctions, of varying paralyses of upper and lower extremities with and without recovery, of successive episodes of transient paralysis, of ascending paralysis of the Landry type and of psychotic episodes. In spite of the striking clinical alterations referable to the nervous system, repeated examinations at autopsy have failed to show any abnormal changes.

My observations, both experimental and clinical, tend to emphasize the functional aspects of the nervous disorder and to suggest the hypothesis that the porphyrins may produce a block in neuromuscular transmission of varying degrees of severity, which may possibly interfere with one or more of the phases of the action of acetylcholine on neurons, synapses, parasympathetic nerve endings and myoneural junctions and in the blood. In addition, the action of the porphyrins on the blood vessels, organs and tissues of the body, as reported here and more extensively in the literature, must be considered.

By way of criticism, it may be pointed out that the use of acetylcholine (or mecholyl) is dangerous and may be productive of more harm than benefit. Greater experience in the use of neostigmine in the symptomatic treatment of this condition is necessary in order to evaluate its therapeutic efficiency.

16. Nachmansohn, D., and Machado, A. L.: The Formation of Acetyl Choline: A New Enzyme, "Choline Acetylase," J. Neurophysiol. **6**:397-403 (Sept.-Nov.) 1943. Nachmansohn, D.; Cox, R. T.; Coates, C. W., and Machado, A. L.: Action Potential and Enzyme Activity in the Electric Organ of Electrophorus Electricus: II. Phosphocreatine as Energy Source of the Action Potential, ibid. **6**:383-396 (Sept.-Nov.) 1943.

SUMMARY

An unusual case of acute porphyria was observed, and striking clinical and laboratory features as well as significant observations at autopsy were studied.

Observations on the status of the capillaries of the skin during the acute attack were made, indicating extreme capillary spasm and evidence of injury to some of the capillaries in the form of aneurysmal dilatation. In addition, definite alterations in gastric contractility were recorded.

The action of injections of acetyl-beta-methylcholine chloride (mecholyl) and neostigmine bromide on the status of the capillaries of the skin, on gastric contractions and on altering the clinical picture was observed, as was the duration of the action.

The use of neostigmine and mecholyl (particularly of the latter) in the symptomatic treatment of the acute phase of this disease may be dangerous and harmful.

RELATION OF NUTRITIONAL DEFICIENCY TO CARDIAC DYSFUNCTION

MAXWELL M. WINTROBE, M.D.
SALT LAKE CITY

That nutritional deficiency is associated with cardiac dysfunction has been recognized for almost sixty years. *Kakke,* which means "disease of the legs," was the subject of a paper by the Japanese author Takaki in 1886.[1] To others this disorder was known as beriberi. The Hollander Eijkman [2] and the Englishmen Fraser and Stanton [3] offered proof of the relation of beriberi to the consumption of a diet of polished rice. The elucidation of the cause of oriental beriberi is correlated with the development of knowledge concerning the vitamins.

HISTORY AND CLINICAL DESCRIPTIONS OF BERIBERI

As is well known, beriberi was prevalent in the Philippines. Chamberlain [4] reported that among 5,000 Philippine Scouts there were always 100 to 600 incapacitated from beriberi. Their diet consisted essentially of 12 ounces (340 Gm.) of beef, 8 ounces (230 Gm.) of white flour, 8 ounces of potatoes or onions and 20 ounces (560 Gm.) of polished rice. On changing the diet by substituting 16 ounces (450 Gm.) of unpolished rice and 1⅗ ounces (45 Gm.) of beans for the 20 ounces of polished rice and by including 20 ounces of sweet potatoes, the number of cases of the disease soon decreased to 50, then to 3, then to 2 and then to none.

Beriberi was also well known in the West Indies and in South America. In Newfoundland, where the cheapest food is white flour and where hunting and fishing in the winter are prohibited by the severity of the weather, the condition makes its appearance in the spring after a winter of a diet consisting mainly of white flour and tea. Not many cases have been described in the United States. In 1928, however, Scott and Herrmann [5] reported *maladie des jambes* in Louisiana. This was observed in the rice-producing parishes of that state among the rice-eating natives, who raised chickens, eggs and other foods but sold them, together with their unpolished rice, and bought with the funds so obtained polished rice. Their favorite and regular diet, *riz et sauce,* was composed of polished rice and bacon grease. These investigators also reported an outbreak of beriberi among the inmates of the Orleans Parish Prison, whose prison fare consisted of a stew of salt pork or beef and boiled hard cowpeas or beans or of carrots and soup and white bread, molasses and water. Milk or fresh vegetables or fruits were not served.

The outstanding symptoms in these persons were palpitation, dyspnea and edema, which led to a diagnosis of heart disease with congestive heart failure. In the rice belt the condition showed a predilection for men. When severe it ran a relatively short course, ending fatally within a few weeks. In the majority of cases the progress was slow. Just preceding or accompanying the appearance of edema of the lower extremities, palpitation developed. Dyspnea, at first barely noticeable, became extremely severe in time. Enlargement of the area of the heart, cardiac dilatation, valvular incompetence, a feeble, rapid soft and irregular pulse and often also an enlarged tender liver, accumulating edema, ascites, hydrothorax and hydropericardium ensued. These signs and symptoms

From the Department of Internal Medicine of the University of Utah.

Presented as guest speaker, at the Fifteenth Annual Postgraduate Symposium on Heart Disease, San Francisco, Oct. 27, 1944, unde rthe auspices of the Heart Committee of the San Francisco Tuberculosis Association. The experimental studies on pigs described in this report were carried out with the aid of grants received chiefly from the Rockefeller Foundation, Parke, Davis & Company and the Upjohn Company.

1. Takaki, K.: Results of the Preventive Measures taken Against the Occurrence of Kakke Among the Japanese Marine Prisoners, Sei-i-Kwai M. J. **41**:43, 1886; cited by Brandon, W. L.: The Cause and Prevention of Beriberi, London, Rebman Co., 1907.

2. Eijkman, C.: Polyneuritis and Beri-Beri, Arch. f. Schiffs- u. Tropen-Hyg. **15**:698, 1911.

3. Fraser, H., and Stanton, A. T.: Studies from the Institute for Medical Research in the Federated Malay States, no. 10, 1909; no. 12, 1911; The Etiology of Beriberi, Lancet **1**:451, 1909; **2**:1755, 1910.

4. Chamberlain, W. P.: Prevention of Beriberi Among Philippine Scouts by Means of Modification in Diet, J. A. M. A. **64**:1215 (April 10) 1915.

5. Scott, L. C., and Herrmann, G. R.: Beriberi ("Maladie des Jambes") in Louisiana, J. A. M. A. **90**: 2083 (June 30) 1928.

were accompanied with a feeling of weakness of the limbs and localized pains and tenderness in the muscles of the calf. Efforts to extend the foot against opposition were weak, and the strength of the grip of the hand was diminished. Sensory disturbances and diminution or absence of deep tendon reflexes completed the clinical picture. Frequently a history of nausea and vomiting was elicited; diarrhea occurred in a number of these persons, and in many pain on pressure in the epigastrium was noted. A presystolic gallop rhythm was observed occasionally. Electrocardiograms revealed small complexes, normal T waves in lead I or III, slight to moderate left ventricular preponderance and some slurring and slight aberrations in the ventricular complexes.

Keefer,[6] working in the Orient, described four varieties of beriberi: namely, (1) neuritic, (2) edematous, (3) cardiac and (4) mixed. A number of reports have appeared in past years describing cases of beriberi in various parts of the world. Among these should be mentioned those of Soma Weiss[7] and his associates in Boston. These workers described beriberi heart as occurring as often as once in every 160 admissions to the medical wards of the Boston City Hospital and as being more frequent than congenital heart disease and subacute bacterial endocarditis. Most of these patients were chronic alcoholic addicts. The cardiovascular disturbances were attributed to thiamine deficiency. They found that the circulatory failure in wet beriberi might involve either the right or the left side of the heart or both. Whereas Aalsmeer and Wenckebach described only "right-sided failure," Weiss also observed patients with orthopnea, cardiac asthma, pulmonary congestion and edema. Breathlessness, palpitation, tachycardia, gallop rhythm, murmurs and venous congestion were present. In severe involvement bounding pulsation of the arteries with pistol shot sounds developed.

Physiologic studies on patients wtih wet beriberi, particularly those with high venous pressures and edema, indicated that the arterioles were dilated, the utilization of oxygen in the venous blood was small and the velocity of blood flow was absolutely or relatively rapid. The vital capacity of the lungs was often severely reduced.

Dock[8] described 5 cases of congestive heart failure of obscure cause, in which there were recurring periods of dyspnea as well as embolic accidents involving the systemic and pulmonary circuits. In these cases pronounced cardiac hypertrophy and mural thrombi in the auricles and ventricles were found, but there was no evidence of endocarditis, myocarditis, coronary disease or arteriolar sclerosis. The diets of these persons had been poor. One of them recovered when fed a "high calory, high vitamin" diet but died in a later episode when he was given a "low salt, cardiac diet." A somewhat similar case was the subject of a clinical pathologic conference at Stanford University[9] and was discussed by Dr. Tinsley Harrison. In this case the heart failure was chronic; it was resistant to digitalis and, peculiarly in view of the alleged causation, also unresponsive to thiamine.

Aalsmeer and Wenckebach[10] ascribed the cardiac enlargement found in patients with wet beriberi to edema of the cardiac musculature. Newcomb,[11] on the other hand, could not demonstrate any increase in fluid and considered the cardiac enlargement to be true hypertrophy. Weiss stated that postmortem studies revealed a moderately or severely dilated heart without coronary or endocardial disease. In some cases of fatal circulatory collapse the cardiac dilatation was lacking. In certain instances myocardial hypertrophy was observed. Microscopically, interstitial edema, or collagen, and hydropic degeneration as reported by Wenckebach[10] was noted, but in other cases no microscopic changes were found. In 2 of Dock's cases[8] small patches of fibrous scarring of the left ventricle were described. In the case of Rytand and Cox,[9] there was no evidence of recent muscular injury, and most of the myocardium was free of scars. Here and there a small dense scar contained a group of lymphocytes.

CAUSE OF BERIBERI

That these cases represent the effects of nutritional deficiency is suggested by strong circumstantial evidence. The relation of dietary deficiency to beriberi has by no means been readily accepted, however. McCarrison,[12] for example, suggested that the disease may have a bacterial

6. Keefer, C. S.: The Beriberi Heart, Arch. Int. Med. **45**:1 (Jan.) 1930.

7. Weiss, S.: Occidental Beriberi with Cardiovascular Manifestations, J. A. M. A. **115**:832 (Sept. 7) 1940.

8. Dock, W.: Marked Cardiac Hypertrophy and Mural Thrombosis in the Ventricles in Beriberi Heart, Tr. A. Am. Physicians **55**:61, 1940.

9. Rytand, D., and Cox, A. J.: Clinical Pathological Conference, Stanford M. Bull. **2**:86, 1944.

10. Aalsmeer, W. C., and Wenckebach, K. F.: Herz und Kreislauf bei der Beriberi-Krankheit, Wien. Arch. f. inn. Med. **16**:193, 1929.

11. Newcomb, C.: Water Content of Heart Muscle in Beriberi Columbarum, Indian J. M. Research **17**:721, 1930.

12. McCarrison, R.: Pathogenesis of Deficiency Disease, Indian J. M. Research **6**:275, 1919.

causation. In one review on the subject [13] as many as twenty-two different theories were advanced to account for it. In time the weight of evidence has come to favor those who hold to the nutritional causation. Even if one accepts this, however, it is necessary to demonstrate the exact nature of the deficiency. A diet composed chiefly of polished rice or the poor diet of the person with chronic alcoholism is lacking in many dietary essentials. Certainly the protein and the vitamin content of such diets are low. Many writers have attributed the edema to protein deficiency. With the recognition of vitamin B and later of a thermolabile fraction, vitamin B_1, the manifestations of beriberi came to be attributed to lack of this dietary essential. As the multiplicity of the vitamins of the B complex has come to be recognized, it has become apparent that beriberi is a multiple nutritional deficiency. I have offered evidence that the nerve degeneration which may occur in this condition may not be due to lack of thiamine.[14] My remarks here will be confined, however, to the cardiac aspects of beriberi.

Weiss,[7] as well as a number of other writers, has described the rapid improvement occurring in cases of wet beriberi when thiamine is given to the patients. This, it is claimed, is much more dramatic than the changes which follow simple rest in bed or rest supplemented by treatment with digitalis and mercurial diuretics.

The etiologic role of thiamine in relation to the wet form of beriberi would be more convincing, however, if it were supported by experimental evidence. Experiments on human beings are naturally few. In those carried out by Williams and his co-workers [15] neither acute, severe deprivation nor moderate, prolonged deprivation of thiamine produced the classic syndrome of beriberi. Minor electrocardiographic changes were observed in a few of the subjects; low blood pressure and vasomotor instability were noted in all. Obviously the extent to which a deliberately produced deficiency can be carried in human beings is limited. One limitation may be production of thiamine by bacteria growing in the intestines and absorption of the vitamin from the bowel, as the studies of Najjar and Holt [16] suggest.

EXPERIMENTAL THIAMINE DEFICIENCY IN ANIMALS

Current views as to the effects of thiamine deficiency in animals are based chiefly on studies in which animals were fed yeast which had been autoclaved or was otherwise treated with the object of destroying thiamine. Such experiments depended on the concept that vitamin B consists of only two parts, the thermolabile and the thermostable, and that by autoclaving the first of these is destroyed. When thiamine was discovered, it was assumed that it was the thermolabile B vitamin. It is now well known that the vitamin B complex consists of ten or more different factors. There is some evidence that in autoclaving yeast more than thiamine is destroyed.[14] It does not follow, therefore, that symptoms and signs developing in animals fed autoclaved yeast are necessarily due to lack of thiamine.

My own studies in thiamine deficiency were not attempted until most of the newer B complex vitamins were available in crystalline form and until I had shown that excellent growth and development take place in swine when they are given a basal diet consisting of casein, sugar, lard and minerals, supplemented with eight crystalline B vitamins and cod liver oil.[17] By failing to give one of the B vitamins or by giving only subminimal amounts, it has been possible to produce deficiency of thiamine or of riboflavin or nicotinic acid or pyridoxine or pantothenic acid. The effects of deficiencies of these vitamins in swine have been described in papers from this laboratory. It has been possible to demonstrate well defined cardiac changes in pigs given inadequate amounts of thiamine.[18]

The outstanding symptoms of acute thiamine deficiency in pigs have been failure of appetite,

13. Vaughan, V. C.: Beriberi, in Vaughan, H. F., and Palmer, G. T.: Epidemiology and Public Health, St. Louis, C. V. Mosby Company, 1922, vol. 2, p. 65.

14. Wintrobe, M. M.; Follis, R. H., Jr.; Humphreys, S.; Stein, H. J., and Lauritsen, M.: Absence of Nerve Degeneration in Chronic Thiamine Deficiency in Pigs, J. Nutrition **28**:283, 1944.

15. Williams, R. D.; Mason, H. L.; Smith, B. F., and Wilder, R. M.: Induced Thiamine (Vitamin B_1) Deficiency and the Thiamine Requirement of Man: Further Observations, Arch. Int. Med. **69**:721 (May) 1942. Williams, R. D.; Mason, H. L.; Power, M. H., and Wilder, R. M.: Induced Thiamine (Vitamin B_1) Deficiency in Man: Relation of Depletion of Thiamine to Development of Biochemical Defect and of Polyneuropathy, ibid. **71**:38 (Jan.) 1943.

16. Najjar, V. A., and Holt, L. E., Jr.: Biosynthesis of Thiamine in Man and Its Implications in Human Nutrition, J. A. M. A. **123**:683 (Nov. 13) 1943.

17. Wintrobe, M. M.; Miller, M. H.; Follis, R. H., Jr.; Stein, H. J.; Mushatt, C., and Humphreys, S.: Sensory Neuron Degeneration in Pigs, J. Nutrition **24**:345, 1942.

18. (*a*) Wintrobe, M. M.; Stein, H. J.; Miller, M. H.; Follis, R. H., Jr.; Najjar, V. A., and Humphreys, S.: A Study of Thiamine Deficiency in Pigs, Bull. Johns Hopkins Hosp. **71**:141, 1942. (*b*) Follis, R. H., Jr.; Miller, M. H.; Wintrobe, M. M., and Stein, H. J.: Development of Myocardial Necrosis and Absence of Nerve Degeneration in Thiamine Deficiency in Pigs, Am. J. Path. **19**:341, 1943. Wintrobe, M. M.; Alcayaga, R.; Humphreys, S., and Follis, R. H., Jr.: Electrocardiographic Changes Associated with Thiamine Deficiency in Pigs, Bull. Johns Hopkins Hosp. **73**:169, 1943.

vomiting and sudden death. Poor growth was a less striking sign. A more or less chronic deficiency was produced in a number of animals by giving subminimal amounts. In such animals I observed weakness, shortness of breath and cyanosis. When in this state, the animals were reluctant to walk about, although obviously there was no paralysis. Examination of 1 such animal revealed the heart beat to be at a rate of 110 per minute, which is somewhat slow for a pig, but the sounds were of good quality, and no murmurs were heard. No rales were found in the lungs. This animal became cyanotic and staggered, fell, struggled stuporously to its feet, fell again on its side and lost consciousness. Just prior to death several convulsive movements were observed.

Another animal in a similar condition after a partial thiamine deficiency of two hundred and forty-six days' duration was reluctant to move about, and soon after he was exercised dyspnea became evident and audible. Some cyanosis was noted, and in time this became pronounced. It was especially noticeable in the ears, the snout and the lips. As time went on, it became more and more difficult for the animal to move about, and finally, after about forty-five minutes, it collapsed after a brief series of jerky movements in the hind quarters. When the animal recovered, it was exercised again. Exercise was followed again by collapse. Convulsive movements and finally death occurred one hour after exercise had been started. Cyanosis and dyspnea had become extreme by this time.

Animals fed inadequate amounts of thiamine have been studied carefully with the aid of electrocardiographic apparatus. Studies of the electrocardiogram of normal pigs show the heart rate to range between 130 and 150 beats per minute, although it may occasionally be as low as 100 and rarely as rapid as 160. The rhythm in my animals was sinuauricular with slight or moderate sinus arrhythmia. Normal electrocardiograms or a moderate tendency to left axis deviation was observed, and only once was a right axis deviation recorded. The PR interval ranged between six hundredths and twelve hundredths of a second. The P waves as measured in lead II were usually four hundredths to six hundredths of a second in duration and 1 to 2.2 mm. in voltage. The duration of the QRS interval was four hundredths to six hundredths of a second, rarely eight hundredths. The T waves varied considerably. In general the T wave in lead I was more often inverted than upright, whereas in the other leads the T wave was usually upright.

In thiamine-deficient pigs two outstanding abnormalities were noted: namely, bradycardia and prolonged conduction time or even dropping of QRS complexes.

It has been claimed that bradycardia results from inanition rather than from the lack of thiamine specifically. We have compared the heart rates of thiamine-deficient pigs with those of animals deficient in other B vitamins. In such animals, the impairment of growth was in many instances much greater than in the thiamine-deficient pigs. We have also made observations on animals given diets that were normal but greatly restricted in amount. In these animals some slowing of the heart rate was observed, but, except in 1 animal subjected to severe dietary restriction, the bradycardia was never as pronounced as in a number of thiamine-deficient pigs.

It is noteworthy that bradycardia was not constant in the thiamine-deficient pigs. Tachycardia occurred when they became excited, after the injection of atropine and when acute cardiac failure developed in association with thiamine deficiency.

Prolongation of auricular-ventricular conduction time was observed in a number of animals. In some there was only first degree auricular-ventricular block, with PR intervals ranging from thirteen hundredths to twenty-eight hundredths of a second. In other animals the ventricular complex was dropped (second degree auricular-ventricular block). The combination of bradycardia and dropping of QRS complexes sometimes caused extreme prolongation of the PR interval, even to two and sixty-eight hundredths seconds. For this animal, nine days later the record suggested auricular fibrillation. This reverted to a normal rhythm without treatment.

Other abnormalities occurred less frequently. In several animals the P waves, especially the P wave in lead II, became abnormally high and broad and sometimes were notched. Occasionally slight widening of the QRS complex was observed, and in a few animals there was a definite shift of the axis to the left. In 1 such animal nodal premature beats were observed. The normal variations in T waves make it difficult to evaluate changes in T waves in thiamine-deficient pigs. The most constant abnormalities appeared to be inversion of the T wave in lead IV.

The observations of my colleagues and me have indicated that the electrocardiographic changes observed in the hearts of these animals were influenced by the degree of thiamine deficiency. With severe deficiency in 1 animal, for example, bradycardia developed as well as complete block

with ectopic ventricular rhythm. Frequent ventricular extrasystoles arising from multiple foci appeared. These abnormalities disappeared after administration of thiamine.

The injection of atropine sulfate into thiamine-deficient animals caused an increase in heart rate and improvement in auricular-ventricular conduction with reappearance of regular QRS complexes.

Pathologic changes in pigs dying in a state of cardiac failure have been distinct. In some of the animals the hearts appeared to be larger than they should have been for the size of the animal. This enlargement seemed to be due both to dilatation and to hypertrophy. In a few a small amount of serous fluid was found in the pericardial cavity. In certain animals pulmonary edema and pleural and peritoneal effusions were found as well. Microscopic examination revealed foci of necrosis in the auricular and ventricular musculature as well as in the cardiac septums. The lesions consisted of focal or diffuse necrosis of myocardial fibers. Such areas were infiltrated by leukocytes.

In animals dying soon after deprivation of thiamine, lesions were found in the auricles and not in the ventricles. In animals dying after longer periods of deficiency, lesions were observed in the ventricles as well as in the auricles. In no instances were pathologic changes found in the ventricles when they were lacking in the auricles.

In 2 animals in which episodes of thiamine deficiency had been observed at intervals during the course of the experiments, stains for connective tissue revealed scars indicative of former necrosis of the myocardium. It is noteworthy, however, that such scars were not always found in pigs which had survived through previous episodes of severe deficiency. It seems, therefore, that, while scars marking the sites of previous myocardial necroses may be found in some animals, the lack of such scars does not necessarily indicate that thiamine deficiency has not been present.

It might be pointed out here that the vagus nerves in these thiamine-deficient animals were normal. At one time it was suggested that the cardiac manifestations of beriberi are due to degeneration of the vagus nerves.

COMMENT

These studies of thiamine deficiency in pigs show conclusively, I believe, that a lack of thiamine produces serious effects on the heart. It does not necessarily follow that the same effects are produced in human beings. There is much evidence that striking differences exist in regard to vitamin requirements for different species and even in regard to the type of abnormality produced in different species of animals by a deficiency of the same vitamin.

The clinical observations in beriberi, however, would make it seem likely that much of what has been observed in thiamine-deficient pigs with reference to the heart is applicable to human beings. Certainly in fatal cases of cardiac failure in which thiamine deficiency is suspected, careful examination of the musculature of the heart, particularly of the auricles, should be made. It should be pointed out that it is to be expected that in some instances of beriberi no lesions will be found. Judging by what is known of the function of thiamine, it is to be expected that a metabolic disorder of the myocardium may occur and may lead to dysfunction and that death may ensue before recognizable lesions have developed.

The evidence at hand suggests that thiamine deficiency impairs the function of the heart by reducing the energy available from the normal processes of carbohydrate metabolism in which thiamine plays a role. Bradycardia is associated with the altered metabolic conditions of thiamine deficiency. Tachycardia is probably the expression of a decompensated heart and also, perhaps, the consequence of vasodilatation and reduced peripheral resistance. Finally serous effusions develop, and all the signs of cardiac failure appear. The evidence at hand also suggests that in thiamine deficiency vagal overaction is present.

The lesions we have found in thiamine-deficient pigs are unlike those that have been described as occurring in the beriberi heart. They suggest instead the types of cases which have been classified as isolated myocarditis, or Fiedler's myocarditis. It is interesting to speculate whether in some instances these may have been examples of thiamine deficiency.

There is no agreement regarding the frequency with which heart failure due to thiamine deficiency may be encountered in the United States. Whereas Weiss observed many cases, others in Boston have recognized few. Besides the cases of acute failure, examples of chronic failure due to thiamine deficiency in which irreversible changes have occurred were reported from San Francisco. I have seen no convincing cases of the latter type and have recognized no more than 1 or 2 of the former. The recognition in human beings of cardiac failure resulting from thiamine deficiency would be greatly simplified if laboratory methods were available which could be relied on as indicative of such deficiency. Many workers, including me, have sought to attain this object, without the desired success. In my animals, thiamine excretion and thiamine

tolerance tests have been employed; the content of pyruvic acid in the blood has been measured, and pyruvic acid tolerance tests have been carried out.[18a] There are difficulties as yet, however, in the application of these procedures to the diagnosis of thiamine deficiency in human beings.

SUMMARY

A number of reports concerning the cardiovascular manifestations of beriberi were studied, and the experimental evidence for the assumption that thiamine deficiency leads to impairment of cardiac function was assembled. In particular, recent studies of thiamine deficiency in pigs were considered. In these animals thiamine deficiency leads to profound changes in cardiac function and produces well defined anatomic lesions. Further investigation is required to demonstrate the role of thiamine in respect to the heart in human beings.

EXTRAGENITAL CHORIONEPITHELIOMA IN THE MALE

HYMAN M. CHERNOFF, M.D.
Resident in Medicine, Grace Hospital

THEODORE S. EVANS, M.D.
Attending Physician in Medicine, Grace Hospital
AND
CHARLES J. BARTLETT, M.D.
Director, Pathologic Laboratories, Grace Hospital
NEW HAVEN, CONN.

Of the malignant tumors occurring in the male, chorionepithelioma is considered to be rare. Some 200 cases have been recorded thus far in the entire literature. Its mode of origin is still uncertain, and many theories have been advanced to explain its presence in the male. These theories have recently been reviewed in a critical survey by Mathieu and Robertson.[1] Most authors accept the hypothesis advanced by Schlagenhaufer,[2] that chorionepithelioma arises in a teratoma in which some structure having the morphologic value of an included ovum had differentiated into trophoblastic epithelium. Theoretically, teratomas may occur anywhere in the body, and they have been described in such areas as the mediastinum, the abdomen, the jaw and the sacrum. If chorionepitheliomas are to be considered as developing from teratomas, they need not always arise in the testes, although the tumors usually are found in these organs.

The existence in the male of chorionepithelioma primary in structures other than the testes has been doubted by such an authority as Prym.[3] However, at least 5 such instances of extragenital origin have been reported in the literature up to 1945.[4] Others have reported cases in which the tumor was considered to be extragenital in origin but in which gross or microscopic examination of the testes was inadequate to exclude the possibility of a primary growth in these organs.[5] Only examination by means of serial section of the testes will eliminate the possibility that a primary growth in a testis was overlooked.

The following case is reported as further evidence of the existence of extragenital chorionepithelioma in the male.

E. C., a white man aged 30, was first admitted to Grace Hospital ward medical service on June 6, 1944. His chief complaints were pain in the left lumbar region of four months' duration and hemoptysis of two weeks' duration.

From the Departments of Medicine and Pathology, Grace Hospital.

1. Mathieu, A., and Robertson, T. D.: Teratomatous Chorionepithelioma in Female and Male: Critical Study of Literature for Years 1935, 1936, 1937; Report of Two Cases in Male, Internat. Abstr. Surg. **64**:158-175, 1939; in Surg., Gynec. & Obst., August 1939.

2. Schlagenhaufer, F.: Ueber das Vorkommen chorionepitheliom- und traubenmolenartiger Wucherungen in Teratomen, Wien. klin. Wchnschr. **15**:571-580, 1902.

3. Prym, P.: Zur Frage der extragenitalen Chorionepithelioma beim Manne, Centralbl. f. allg. Path. u. path. Anat. **49**:98-101 (Aug.) 1930.

4. Erdmann, J. F.; Brown, H. A., and Shaw, H. W.: Chorionepithelioma in Male of Extragenital Origin, Urol. & Cutan. Rev. **45**:1-6 (Jan.) 1941. Fenster, E.: Ueber ein extragenitales Chorionepitheliom beim Manne mit positiver Hypophysen vorderlappenreaktion, Frankfurt. Ztschr. f. Path. **46**:403, 1934. Gerber, I. E.: Ectopic Chorionepithelioma, J. Mt. Sinai Hosp. **2**:135-142 (Sept.-Oct.) 1935. Kantrowitz, A. R.: Extragenital Chorionepithelioma in a Man, Arch. Path. **13**: 186-188 (Jan.) 1932. Weinberg, T.: Primary Chorionepithelioma of Urinary Bladder in Male: Report of a Case, Am. J. Path. **15**:783-796 (Nov.) 1939.

5. Arendt, J.: Das Chorionepitheliom des Mannes, Fortschr. a. d. Geb. d. Röntgenstrahlen **43**:728-735 (June) 1931. Bonn, H. K., and Evans, N.: Extragenital Chorionepithelioma in Male with Associated Gynecomastia: Report of a Case, Am. J. Surg. **58**:125-132 (Oct.) 1942. Bonney, V.: On Chorionepithelioma of Congenital Origin, Arch. Middlesex Hosp., London **7**: 88-106, 1906. Heaney, H. G.: Extragenital Chorionepithelioma in Male, Am. J. Cancer **19**:22-30 (Sept.) 1933. Krassnianskia, P. V.: Causes of Chorionepithelioma in Men Outside of Sexual Sphere, Mosk. M. J. **9**: 1-7, 1929. Lambert, S.W., and Knox, L.C.: Intrathoracic Teratomata, Tr. A. Am. Physicians **35**:17-62, 1920. Miller, J., and Browne, F. J.: Extragenital Chorionepitheliomata of Congenital Origin (with Report of a New Case of Chorionepithelioma in a Male), J. Obst. & Gynaec. Brit. Emp., **29**:48-67, 1922. Ritchie, J.: A Case of Embryoma Occurring in the Mediastinum, J. Obst. & Gynaec. Brit. Emp. **4**:65-73, 1903. Rottino, A., and DeBellis, H.: Extragenital Chorioma: Its Relation to Teratoid Vestiges in the Testicles, Arch. Path. **37**: 78-80 (Jan.) 1944. Weber, F. P.: Malignant Chorionepithelioma in a Male: Fatal Intraperitoneal Hemorrhage from Rupture of a Metastatic Growth in the Liver, Practitioner **101**:31-36, 1918. Mathieu and Robertson.[1]

The patient had been in good health until one year prior to his admission, when he first noted slight swelling of the breasts lasting about one week and then subsiding spontaneously. Six months before his entry into the hospital, he experienced a dragging sensation in the left testis, at which time examination by his family physician disclosed a varicocele, for which a suspensory was prescribed. Persistent aching pain in the left lumbar region developed about four months before his admission to the hospital. Over this period there was a progressive loss of weight of 18 pounds (8.2 Kg.). Two weeks prior to his entry a cough productive of blood-streaked sputum developed. At this time he also noted enlargement and tenderness of both breasts.

Physical examination revealed a pale, well developed, somewhat emaciated white man weighing 136 pounds (62.5 Kg.). Both breasts were enlarged and tender, with prominent areolas and Montgomery follicles. No increase in polymorphonuclear neutrophils. The reaction to the cephalin-cholesterol flocculation test was 2 plus. Wassermann and Kahn reactions of the blood were negative. Examination of the sputum revealed the presence of blood and a mixed flora of gram-positive and gram-negative cocci and bacilli; no tubercle bacilli were found. The reaction to the Friedman test of the blood and of the spinal fluid for presence of gonadotropic hormone was strongly positive.

Roentgenographic examination revealed the presence of several large rounded and well defined opaque masses in both pulmonary fields. Examination by intravenous pyelography demonstrated displacement of the left kidney laterally and forward by a large mass in the preaortic area. The series of gastrointestinal roentgenograms revealed displacement of the stomach downward, forward and laterally by a mass in the prevertebral area. The transverse colon and the splenic flexure were also displaced downward.

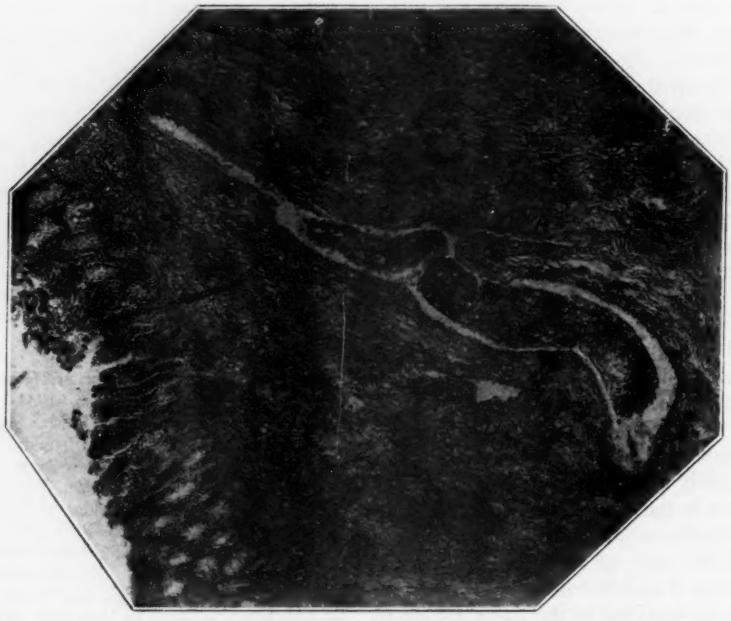


Fig. 1.—Chorionepithelioma; group of tumor cells in blood vessel of intestinal wall, (× 35).

colostrum could be expressed. Several small nontender lymph nodes were present in both axillas. The blood pressure was 116 systolic and 60 diastolic. The abdomen was not distended, but definite tenderness was elicited in the left upper quadrant, where a large irregular firm mass was palpable. This mass extended to 3 fingerbreadths below the border of the ribs. It was immovable and seemed fixed to the posterior abdominal wall. No friction rub could be heard in this area. The liver and kidneys were not felt. Moderate tenderness was present in the left lumbar region. There was definite scoliosis of the vertebral column, with convexity directed toward the left. The right testis and epididymis were normal except for decreased sensitivity to pressure. The left testis was smaller and much softer than the right, but no nodules were palpated.

Urinalysis done at the time of the patient's admission to the hospital revealed a trace of albumin. The erythrocyte count was 4,390,000 per cubic millimeter and the leukocyte count 11,800 per cubic millimeter, with a slight

Because of the clinical and roentgenologic findings, a presumptive diagnosis of chorionepithelioma, probably primary in the left testis with metastases to the preaortic lymph nodes and lungs, was made. The left testis was removed through an inguinal incision. Serial sections of the testis were made in an effort to find the primary lesion. Gross examination revealed a very small hemorrhagic area, about 0.80 cm. in diameter, in the lower pole of the testis. Sections through this area, however, showed no tumor growth, revealing merely fibrous tissue and a few pigment-containing cells. The cells lining the spermatic tubules were vacuolated. There was no evidence of spermatogenesis. In sections from different parts of the testis, clusters of polyhedral cells were found which were considered to be groups of interstitial cells. The sections were sent to Dr. Shields Warren, of Boston, who confirmed these observations and who wrote suggesting that they represented atrophy as a result of the output of female sex hormone. In view of the hormonal evidence and the distribution

of metastases, he concluded that the lesion was chorionepithelioma, probably extragenital in origin but, however, developing from germinal epithelium originating in the urogenital ridge.

The patient was discharged to his home on July 7, 1944 and during the next five months was given intensive roentgen ray therapy in the department of radiology of Grace Hospital. Despite the fact that serial roentgenograms of the chest showed a definite decrease in the size of the pulmonary metastatic nodules and although the cough was less distressing, the patient became worse, requiring opiates for relief of the pain in the left lumbar region. Follow-up examinations in the tumor clinic during the course of the roentgen ray therapy revealed some decrease in the size of the mass in the left upper quadrant. Two weeks of occipital headaches accompanied with amblyopia culminated in an episode of sudden loss of consciousness. This was followed by complete amaurosis for a period of five minutes and led to his readmission to the hospital on November 1, five months after his first admission.

His weight at that time was 114 pounds (52.5 Kg.), a further loss of 22 pounds (10 Kg.). Physical examination revealed a cachectic man, dyspneic and somewhat confused. The left pupil was larger than the right but reacted well to light. The fundi and visual fields were normal. Both breasts were enlarged, the right more than the left. The previously described mass in the left upper abdominal quadrant was unchanged. The left testis was absent. The right testis seemed smaller and softer than normal, but no nodules were felt. The erythrocyte count was 2,800,000 per cubic millimeter and the leukocyte count 9,500 per cubic millimeter, with 88 polymorphonuclear neutrophils. The reaction to the cephalin-cholesterol flocculation test was 1 plus. The result of the sulfobromophthalein sodium test for hepatic function was normal. Reaction to the Friedman test of the urine for gonadotropic hormone again was highly positive.

The patient complained of severe cough, dyspnea and lumbar pain and required heavy sedation. He became progressively weaker and died on the tenth day in the hospital.

Autopsy showed the following details:

Gross Examination.—The body showed considerable emaciation. Both breasts were moderately enlarged. No fluid was found within the peritoneal cavity. The stomach and small bowel occupied their usual sites and on gross examination revealed no lesions. An irregular nodular mass, measuring 11 by 7 by 5 cm. was observed to lie anteromedial to the left kidney and below the left adrenal gland at the level of the second and third lumbar vertebrae. The mass was adherent to the periosteum of the vertebral bodies but did not involve the compact bone. It was distinctly separate from the kidney and adrenal gland and did not infiltrate these organs. Anteriorly it was covered with peritoneal reflections, and when these were divided the tumor mass could be dissected free and removed. On cut section dense strands of pale red fibrous tissue surrounding small cystlike spaces and small areas of necrosis were seen to make up the entire structure.

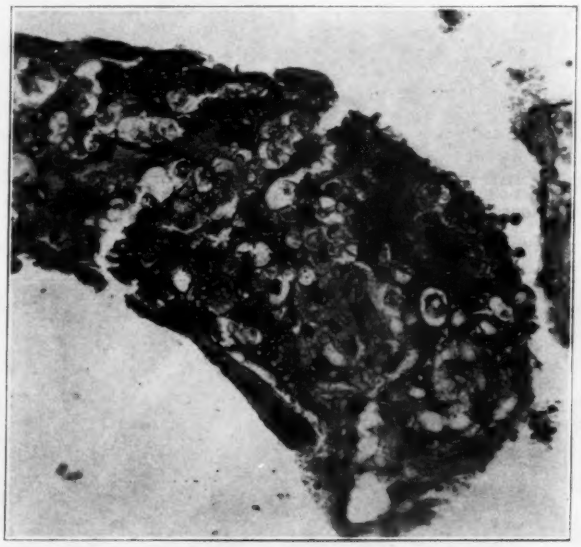


Fig. 2.—Same as figure 1 (high power, × 325).

The kidneys together weighed 295 Gm. When the capsule was stripped from the cortex of the right kidney, a few small dark lesions were seen which on section had the appearance of a metastatic tumor. The left kidney showed no tumor nodules. The renal vessels were free of tumor growth. On cross section, the adrenals showed no abnormalities. The prostate was not enlarged. The left testis was absent. The right testis was small, and on multiple cross section no nodules were encountered. There was no enlargement of the mesenteric, parailiac or inguinal nodes.

The large bowel contained much dark soft material, suggestive of old blood. An irregular dark polypoid mass, about 2 cm. in diameter, was seen arising from the mucosa of the large bowel at the first part of the descending colon. There was no surrounding area of infiltration or necrosis.

The liver weighed 1,340 Gm. On cut section, no gross evidence of malignant infiltration was seen. The spleen weighed 80 Gm. and appeared normal.

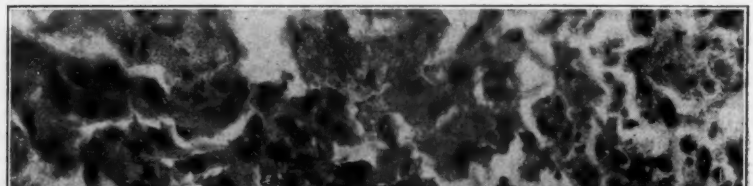


Fig. 3.—Chorionepithelioma, section of small nodule attached to pleura (× 375).

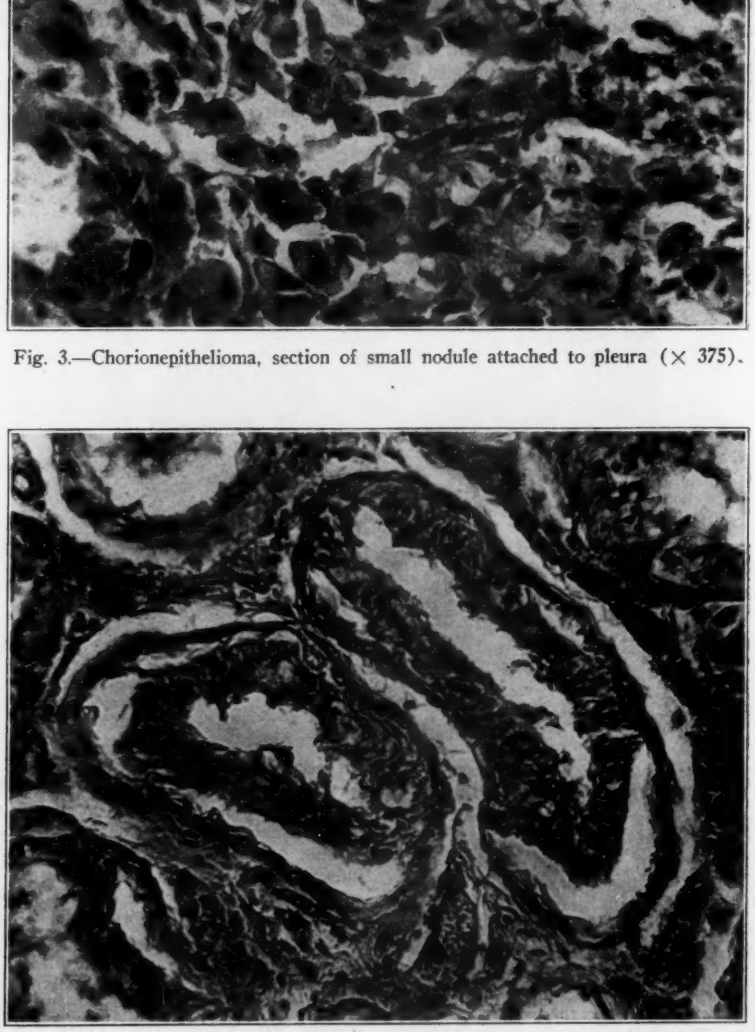


Fig. 4.—Section of testicle, exhibiting tubular atrophy and prominence of interstitial cells (× 300).

About 150 cc. of pink-tinged fluid was found in the left pleural cavity. The right pleural cavity was for the most part obliterated by fibrous adhesions. The right lung weighed 1,120 Gm., and the left lung weighed 1,100 Gm. Both lungs were extensively infiltrated with tumor masses of varying sizes up to 5 cm. in diameter. On cross section, these masses were seen to be variegated in nature and irregular in shape. Some of the tumor masses were firm, homogeneous and yellow; others had a mulberry-like appearance and were filled with soft cystic spaces containing dark hemorrhagic material. Many of the tumor nodules formed hemispherical masses above the visceral pleural surface and in a few areas invaded the pleura and

overlying ribs. The diaphragmatic surface of the right lung was covered with a thick white exudate, which was adherent to the diaphragm. The remaining pulmonary fields showed dense congestion and edema as well as small areas of atelectasis. The trachea and bronchi were free of exudate and obstructing masses. The mediastinal lymph nodes were not enlarged and had the appearance of normal lymphoid tissue. The heart weighed 270 Gm. and presented no abnormalities.

The brain appeared somewhat edematous, and the convolutions were slightly flattened. The meninges were free of exudate. A firm dark red cystic tumor mass was seen to occupy the posterior pole of the right occipital cortex. This mass measured about 3 cm. in its greatest diameter. Multiple transverse sections of the brain showed no other metastatic lesions.

Microscopic Examination.—The retroperitoneal tumor was found to be so completely necrotic that no classification of it could be made. This necrosis may have resulted from the large amount of radiation to which the patient was subjected.

The structure of the tumor, however, was well seen in sections made of the nodules found in the large bowel, the parietal pleura, the lung, the brain and the kidneys. There were much hemorrhage and necrosis in the tumors in all these locations. The tumor cells varied considerably in size and shape. Most of these were large and pale, with large round or oval nuclei. These cells resembled the Langhans cell seen in placental tissue. Scattered among the Langhans cells were groups of syncytial cells with indefinite outline containing small hyperchromatic nuclei. In the section of the tumor removed from the descending colon, clusters of such cells were seen in the lumen of a dilated blood vessel. Sections of the lungs from portions apparently not involved in the tumor growth showed them to be somewhat more fibrous than normal and infiltrated with great numbers of small round cells and plasma cells, indicating a chronic inflammatory process, probably the result of the extensive roentgen ray therapy. No tumor growth was found in the liver, spleen, adrenal glands, or prostate.

Multiple sections of the right testis failed to reveal any tumor tissue. No spermatogenesis was present. There was moderate hyperplasia of the interstitial cells, similar to that observed in the left testis removed at operation.

Pathologic Diagnosis.—The lesions were diagnosed as chorionepitheliomas with their primary site probably in the retroperitoneal mass in the region of the left kidney and with metastases to the lungs, pleura, ribs, kidney, brain and large bowel.

SUMMARY

A case of extragenital chorionepithelioma occurring in a man was encountered.

The clinical picture the patient presented, that of gynecomastia, pain in the lumbar region, hemoptysis, loss of weight and rapid downhill course, is characteristic of tumors of embryonal urogenital origin which have widely metastasized.

The production by the tumor of large amounts of gonadotropic hormone was demonstrated by the highly positive reaction to the Friedman test with both urine and spinal fluid. The enlargement of the breasts, the absence of spermatogenesis and the interstitial hyperplasia noted in the testis are considered to have been produced by the excessive amounts of this hormone.

Intensive roentgen ray therapy over a period of five months did not alter the outcome, although it did produce some symptomatic relief and some decrease in the size of the pulmonary metastases.

Careful histologic examination of both testes revealed no tumor growth. The large retroperitoneal mass demonstrated at autopsy in the region of the left kidney was believed to represent the primary site of the chorionepithelioma.

Sections of several of the metastatic nodules revealed the presence of Langhans and syncytial cells typical of chorionic tissue.

The location of the primary growth suggests its derivation from germinal epithelium in the region of the primitive urogenital anlage.

PLASMA QUINACRINE CONCENTRATION IN TREATMENT OF PLASMODIUM VIVAX MALARIA ACQUIRED IN THE SOUTH PACIFIC

CAPTAIN LESTER D. ELLERBROOK
SANITARY CORPS, ARMY OF THE UNITED STATES

LIEUTENANT COLONEL STUART W. LIPPINCOTT
MEDICAL CORPS, ARMY OF THE UNITED STATES

SECOND LIEUTENANT CARLO F. CATENO
SANITARY CORPS, ARMY OF THE UNITED STATES

AND

MAJOR HARRY H. GORDON

AND

COLONEL ALEXANDER MARBLE
MEDICAL CORPS, ARMY OF THE UNITED STATES

The development of an adequate technic for determining plasma quinacrine concentrations [1] has facilitated the investigation of several important phases of therapy for soldiers evacuated from the South Pacific with chronic relapsing Plasmodium vivax malaria. The concentrations of quinacrine in plasma attained at selected intervals as the result of oral administration of known amounts of the drug have been determined for specific dosages. Determinations were made on 6,733 specimens from 291 patients during and after treatment for four hundred and twelve attacks of malaria at Harmon General Hospital.

METHODS

The method of Brodie and Udenfriend [1] was employed in the determination of the concentration of quinacrine in plasma. Plasma was used instead of whole blood because these investigators stated that variations in values in whole blood quinacrine concentrations are often reflections of changes in the number of leukocytes, since they contain a concentration several hundred times that of plasma. Shortly after the 20 to 25 cc. specimens of oxalated blood were drawn, they were centrifuged at 1,500 rotations per minute for one hour. The plasma was then drawn off carefully so as to minimize contamination with white cells. Most of the determinations were made not immediately but as time permitted, since it was shown that no demonstrable changes in concentration of quinacrine resulted when plasma was refrigerated for as long as seven days or kept in the frozen state for as long as four months.

In the method used quinacrine is determined by measuring its fluorescence in an acid medium. Ethylene dichloride is used to extract quinacrine as the free base from alkaline plasma. The quinacrine base is then extracted from the ethylene dichloride into lactic acid. The intensity of the fluorescence which is produced in this acid phase by ultraviolet rays of the proper wavelength is compared with that produced by a known amount of quinacrine. The measurements are made with the model 12 Coleman Electronic Photofluorometer, the B2 filter being used for the exciting light and the PC9 filter for the emitted light.

Small amounts of fluorescing quinacrine degradation products which are present in the plasma of patients undergoing therapy may accompany the quinacrine in this extraction procedure. Brodie and Udenfriend suggested the removal of these degradation products for precise work by washing the ethylene dichloride extract with 10 per cent sodium hydroxide solution. This alkaline wash was not used for our routine determinations.

Procedure.—1. Ten cubic centimeters of plasma and 3 cc. of phosphate buffer at p_H 8 were extracted with 30 cc. of ethylene dichloride.

2. A 20 cc. portion of the ethylene dichloride layer was extracted with 1 cc. of water and 10 cc. of lactic acid U. S. P. XII (Mallinckrodt).

3. Standards and blanks were prepared in duplicate by additions of 1 cc. portions of quinacrine solutions of known concentrations to 10 cc. portions of lactic acid, each being shaken with 20 cc. of ethylene dichloride.

4. The intensity of fluorescence produced in the lactic acid phase of the plasma extract was compared with that of the standards, and the concentration in micrograms of quinacrine base per liter of plasma was calculated. One gram of quinacrine dihydrochloride dihydrate is equivalent to 0.78 Gm. of quinacrine base. The results presented in all tables and figures are reported as quinacrine base.

Reliability of the Method.—The precision of the results obtained by the method employed may be gaged by the data presented in table 1. An average blank value corresponding to 3 micrograms of quinacrine per liter of plasma was obtained. The blanks were not subtracted from the values obtained. When known amounts of quinacrine were added to 5 or 10 cc. por-

1. Brodie, B. B., and Udenfriend, S.: The Estimation of Atabrine in Biological Fluid and Tissues, J. Biol. Chem. **151**:299-317 (Nov.) 1943.

tions of plasma, the proportions recovered were usually approximately 96 per cent, although on an occasional day all results were either slightly high or slightly low. The average recoveries, the ranges of values and individual results obtained in two consecutive series of such recovery experiments are shown in table 1.

TABLE 1.—*Recoveries of Quinacrine Added to Plasma*

Number of Tests	Micrograms Quinacrine Added to 5 or 10 Cc. Plasma	Average Recovery	Range
49	0	3 micrograms per liter	0 to 4 micrograms per liter
16	0.25	96%	88 to 106%
30	0.50	96%	86 to 110%
49	1.00	96%	82 to 104%

Micrograms of Quinacrine Added to 5 Cc. Plasma	Micrograms of Quinacrine Recovered							
	Series 1				Series 2			
0	0	0	0.01		0.02	0.02	0.015	
0.25	0.22	0.23	0.24	0.24	0.24	0.26	0.26	0.24
0.50	0.48	0.48	0.47	0.48	0.51	0.50	0.50	
1.00	1.00	0.98	0.98	0.98	1.00	1.00	0.98	0.96

Results Obtained in Two Consecutive Series of Determinations

TREATMENT

The patients in this study all received quinacrine dihydrochloride dihydrate (hereafter referred to under treatment as quinacrine hydrochloride) and were divided into four groups. Treatment for patients in groups I, II, and III was begun at the first regular time for drug administration, that is, at meal time or at midnight, after their admission to the ward with an attack of malaria, which was arbitrarily defined as temperature over 100 F. and a smear positive for Plasmodia. The approximate times of the meals were 7:30 a.m., 11:30 a.m. and 5 p.m.

Group I—Standard treatment: Quinacrine hydrochloride, 0.2 Gm., was given orally for five doses with meals and at midnight, totaling 1 Gm. in twenty-four hours of treatment. This was followed by 0.1 Gm. three times daily with meals until a total of 2.8 Gm. had been given.

Group II.—A total of 2.8 Gm. of quinacrine hydrochloride was given as in group I, followed by a twenty-four hour rest period, then 0.01 Gm. of pamaquine naphthoate three times daily for three days followed by 0.1 Gm. of quinacrine hydrochloride six days per week through the sixtieth day from the initial day of treatment.

Group III.—A total of 2.8 Gm. of quinacrine hydrochloride was given as in group I, followed by 0.1 Gm. of quinacrine hydrochloride six days per week from the eighth through the sixtieth day from the initial day of treatment.

Group IV.—The amount of quinacrine hydrochloride given by mouth was 0.4 Gm. at 9 a.m. followed by 0.2 Gm. at 4:30 p.m. and at 1 a.m. Thereafter 0.2 Gm. was given at 9 a.m. and at 9 p.m. for six days The total was 3.2 Gm.

RESULTS

Fasting Plasma Concentrations (Groups I, II and III).—The 104 patients studied included 54 patients of group I and 50 patients of groups II and III. The concentration of quinacrine in plasma obtained before breakfast was determined for all these patients for eight days while undergoing similar therapy. Thereafter the fasting levels were determined for only the 54 patients of group I on the ninth and tenth days, every other day for ten more days and again on the twenty-eighth day.

The average fasting plasma quinacrine concentrations obtained on each full day of treatment of the 104 patients as well as the average values for each treatment group are shown in table 2. It can be seen that with the exception of those obtained on the morning of the first day of treatment the fasting values were but little affected by the variation of the hour at which treatment had been instituted because

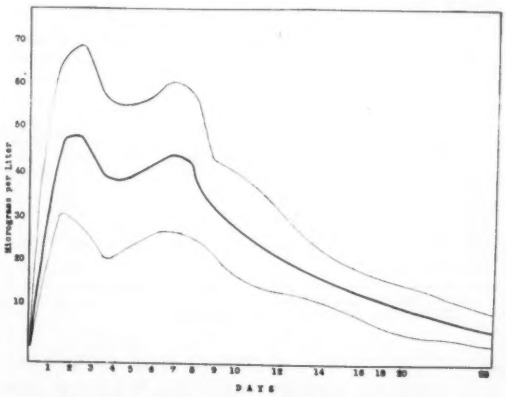


Fig. 1.—Fasting plasma quinacrine concentrations in 104 patients for eight days and in 54 patients thereafter (2.8 Gm. of quinacrine hydrochloride in one week). The heavy line indicates average values. Fine lines indicate 1 standard deviation (includes approximately two thirds of all values).

the total amounts of drug received by a given time were practically identical in each case.

The average values, however, do not necessarily represent the values which would be attained by any given person receiving a similar type of therapy. There was much variation between the results obtained on different persons, although most of the curves were characterized by a rapid rise, a dip and then a slower rise until treatment was discontinued. The extent of these variations may be gaged from chart 1, in which are shown the standard deviations from the average values. The area between the upper and lower curves includes approximately two thirds of all values. One

third of the values were either above or below these limits, and isolated values of as high as 120 and as low as 5 micrograms per liter were obtained during the week of treatment.

After the administration of quinacrine hydrochloride was discontinued, the plasma concentrations fell slowly, so that by four weeks after the attack the average value was 8 micrograms per liter, with a range of 2 to 21 micrograms per liter. From the eighth to the ninth day the average concentration decreased by 21 per cent, and from then until the eighteenth day the daily decreases were approximately 10 per cent. During the next ten days the levels decreased more slowly.

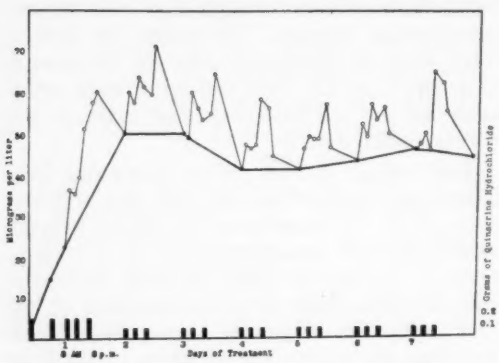


Fig. 2.—Diurnal variation of plasma quinacrine concentrations in 104 patients given 2.8 Gm. of quinacrine hydrochloride.

Diurnal Variations of Plasma Quinacrine Concentration in 104 Patients.—A simultaneous study of the diurnal variations of the plasma concentrations during the week of treatment was made by obtaining blood specimens from six groups of approximately 15 patients each at two or at four hours after administration of the drug at breakfast, dinner and supper respectively in addition to the fasting specimens.

The average rises in values above the individual daily fasting levels on the second through the seventh days were as follows: two and four hours after breakfast, 2.3 and 6.8 micrograms per liter; two and four hours after dinner, 6.6 and 11.3 micrograms per liter, and two and four hours after supper, 12 and 8 micrograms per liter. The average daily changes at the specified times are shown in chart 2, in which these values are superimposed on the average fasting curve for the 104 men. The average patterns on each day are similar, with moderately elevated values being found early in the day and with peak values being found later in the day. The individual curves again showed considerable irregularity, with an occasional postprandial level being slightly below the fasting level.

Plasma Quinacrine Concentrations in Groups 2 and 3.—All specimens from the 215 patients in these two groups were obtained at approximately four hours after breakfast. The total amount of quinacrine hydrochloride given to the patients in group III was approximately 7.3 Gm., while those in group II received 0.4 Gm. less. The average values obtained are shown in chart 3. Again there were considerable individual differences, as shown by the standard deviations. After the dosage of the

TABLE 2.—*Average Fasting Plasma Quinacrine Concentrations with Reference to Dosage*

Day of Attack			Full Days of Treatment													
			1		2		3		4		5		6		7	8
Time of First Treatment	Per Cent of Patients	Drug,† Gm.	Fasting Value*	Drug,† Gm.	Fasting Value*	Drug,† Gm.	Fasting Value*	Drug,† Gm.	Fasting Value*	Drug,† Gm.	Fasting Value*	Drug,† Gm.	Fasting Value*	Drug,† Gm.	Fasting Value*	Fasting Value*
7:30 a. m.	6	0.8	54	1.2	65	1.5	53	1.8	47	2.1	45	2.4	49	2.7	56	2.8	33
11:30 a. m.	22	0.6	35	1.1	53	1.4	49	1.7	44	2.0	44	2.3	48	2.6	52	2.8	48
5:00 p. m.	54	0.4	21	1.0	50	1.3	48	1.6	39	1.9	40	2.2	42	2.5	44	2.8	47
12 midnight.....	18	0.2	15	1.0	52	1.3	56	1.6	43	1.9	44	2.2	45	2.5	51	2.8	54
Average.....		0.4	25	1.0	52	1.3	50	1.6	41	1.9	42	2.2	44	2.5	48	2.8	48

* Micrograms per liter.
† Total drug received by 12 midnight.

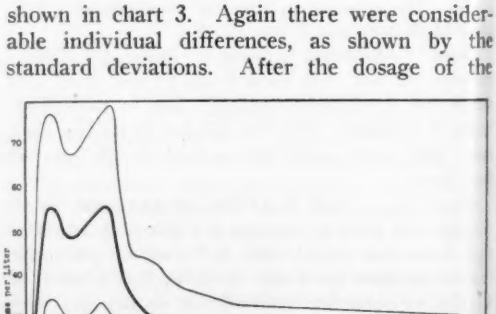


Fig. 3.—Plasma quinacrine concentrations at 11 a.m. of 226 patients who were given 2.8 Gm. of quinacrine hydrochloride in one week. The heavy line indicates average values for hours after administration of 0.1 Gm. Fine lines indicate 1 standard deviation (includes approximately two thirds of all values).

drug was reduced, the levels at first dropped rather sharply and then gradually leveled off to a concentration of approximately 23 micrograms per liter. The rates of fall of the concentrations of groups II and III did not differ significantly. These daily 11 a.m. levels during the week of active treatment exceeded the average fasting levels of the 104 patients (in chart 1) by the amounts shown in table 3 and approximated the values found for the smaller group of 15 patients previously studied at 11 a.m.

TABLE 3.—*Elevations Above Fasting Levels Four Hours After Administration of Quinacrine Hydrochloride with Breakfast*

	Days						
	First	Second	Third	Fourth	Fifth	Sixth	Seventh
	Microgram per Liter	Microgram per Liter	Microgram per Liter	Microgram per Liter	Microgram per Liter	Microgram per Liter	Microgram per Liter
A	9	5	4	7	8	8	8
B	13	7	10	6	9	6	4

A = elevation of 11 a. m. concentrations of 215 patients above fasting concentrations of 104 patients on the same type of therapy.

B = Elevations of 11 a. m. concentrations of 15 patients above their own fasting values on the same days.

When the administration of the drug was stopped, the levels decreased approximately 20 per cent per day for two days and then at a rate of approximately 10 per cent per day until the eighteenth day, at which time the levels were practically identical with those obtained with group 1 at this time (15 micrograms per liter).

Plasma Quinacrine Concentrations in Successive Attacks.—Plasma quinacrine concentrations were determined in successive attacks in 71 patients to observe whether the levels were similar on a repetition of the same dosage (2.8 Gm. in one week). In some instances curves were obtained for three successive attacks. Many of the curves obtained in two or even three attacks resembled each other closely, but there were also instances in which wide differences existed. Chart 5 is an example of two successive curves for 1 patient, with both representing levels considerably higher than the average for group II

TABLE 4.—*Average Fasting Plasma Quinacrine Levels of the Groups Receiving 2.8 Gm. and 3.2 Gm. Respectively*

	Attack		Days of Treatment													
			1		2		3		4		5		6		7	8
Group	Drug by End of Day, Gm.	Microgram per Liter	Drug by End of Day, Gm.	Level*	Gm.†	Level*	Gm.†	Level*	Gm.†	Level*	Gm.†	Level*	Gm.†	Level*	Gm.†	Level*
3.2 Gm. group	0	3	0.8	36	1.2	43	1.6	46	2.0	49	2.4	51	2.8	61	3.2	63
2.8 Gm. group	0.4	25	1.0	51	1.3	51	1.6	42	1.9	42	2.2	44	2.5	47	2.8	48

* Level refers to micrograms per liter.
† Grams of drug received by end of day.

Plasma Quinacrine Concentrations in Group 4.—The specimens of plasma were obtained from these patients before the morning doses of drug (twelve hours after last dose), and, in addition, on days 1, 2 and 7 specimens were obtained at 2 p.m. (five hours after the morning dose). The average concentrations found at these times are shown in chart 4, in which the solid line represents the "fasting" values and the broken lines connect the three five-hour values with the preceding and the following average fasting values. The shape of this fasting curve differs from those of the other groups because the schedules of doses were different. The levels on the first three days tended to be lower because less drug had been given, but by the fourth day the same amount of drug had been received by each group and the fasting concentrations were nearly the same. After this time, however, the values in group IV gradually exceeded the others until by the morning of the eighth day the two figures were 48 and 63 micrograms per liter.

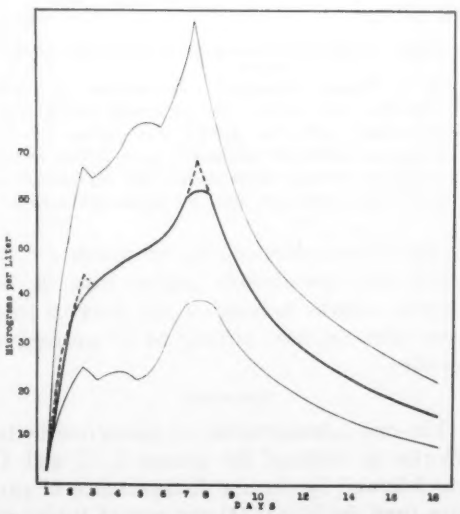


Fig. 4.—Plasma quinacrine concentrations of 35 patients who were given 3.2 Gm. of quinacrine hydhrochloride in one week. The heavy line indicates average fasting values. The broken line indicates five hours after administration of 0.2 Gm. Fine lines indicate 1 standard deviation (includes approximately two thirds of all values).

treatment. In contrast is chart 6, showing three successive curves for 1 patient in whom the levels were consistently below the average for group I and II treatment. Another variant is seen in chart 7, in a patient who broke through suppressive treatment with an attack of malaria after measles had developed. The

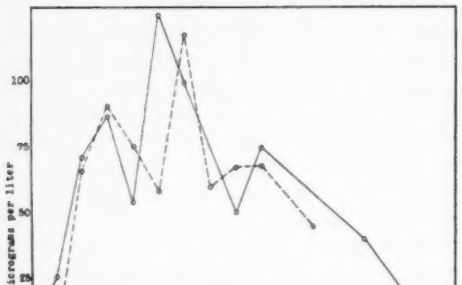


Fig. 5.—Plasma quinacrine concentrations in successive attacks (high levels). The continuous line indicates the nineteenth attack and the broken line the twentieth attack.

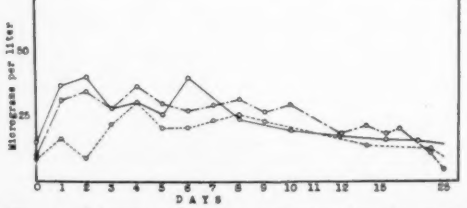


Fig. 6.—Plasma quinacrine concentrations in successive attacks (low levels). The eighteenth attack was a breakthrough while the patient was taking 0.1 Gm. of quinacrine hydrochloride daily. Long dashes indicate the sixteenth attack; short dashes, the seventeenth attack, and the continuous line, the eighteenth attack.

plasma concentration during treatment for this attack was considerably higher than in two previous attacks because of the start of treatment with the level already at 35 micrograms per liter.

COMMENT

The oral administration of quinacrine hydrochloride as outlined for groups I, II and III was followed by prompt disappearance of parasites from the blood; 90 per cent of the smears in two hundred and thirty-four attacks were negative within thirty-two to forty-eight hours after initiation of treatment. There was also prompt subsidence of fever; 66 per cent of the patients in three hundred and thirteen attacks had normal temperatures on the second day after initiation of treatment and 94 per cent by the fifth day. Symptoms were readily relieved, and by seventy-two hours after the beginning of treatment the patients were usually able to leave the ward for recreational purposes.

The reliability of the method for determining plasma quinacrine concentrations is apparent from table 1, in which it is shown that the average recovery is 96 per cent when amounts as small as 0.25 microgram of quinacrine hydrochloride are added to 5 or 10 cc. of plasma. The values obtained for the concentrations in the plasma of patients undergoing therapy are probably slightly higher than the actual concentrations because of the inclusion of small amounts of fluorescent degradation products of quinacrine. While the average values for the fasting and the postprandial curves gave a rather

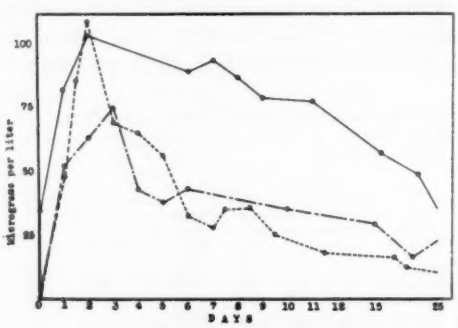


Fig. 7.—Plasma quinacrine concentrations in successive attacks (breakthrough during measles). Measles developed in this patient thirty-five days after completion of treatment with quinacrine hydrochloride, 2.8 Gm., and while he was taking 0.1 Gm. of quinacrine hydrochloride daily. A breakthrough with another attack of malaria occurred at this time, while the plasma quinacrine concentration was 25 micrograms per liter. Short dashes indicate the seventh attack; long dashes, the ninth attack, and the continuous line the tenth attack.

constant pattern, it is to be emphasized that there were many individual variations. When group I treatment was discontinued, the levels fell rather slowly, but when this was followed by 0.1 Gm. of quinacrine hydrochloride six days per week the levels fell more slowly and did not fall below a concentration of approximately 23 micrograms per liter. At this average concentration there was only an occasional breakthrough with another attack.

The clinical effectiveness of the administration of 2.8 Gm. of quinacrine hydrochloride in one week is obvious, but it may also be true that a smaller amount might be effective. For example, while this dosage gives average fasting levels from the second to the eighth days

of 41 to 52 micrograms per liter, a lower level might conceivably be adequate. We have had no experience with a lower dosage, although some of our patients in whom the concentrations were lower than the average had satisfactorily prompt relief from symptoms.

SUMMARY AND CONCLUSIONS

The concentration of quinacrine was determined in 6,733 specimens of plasma obtained from 291 patients both during and after four hundred and twelve attacks of P. vivax malaria of South Pacific origin. The standard treatment was the oral administration of 2.8 Gm. of quinacrine hydrochloride in approximately seven days. For 33 patients, however, the amount was 3.2 Gm.

The range of the daily average fasting values on the standard 2.8 Gm. treatment from the second through the eighth day was 41 to 52 micrograms per liter, while the concentrations of specimens taken four hours after the first daily dose of drug were 49 to 57 micrograms per liter. After the administration of the drug was discontinued, the levels decreased slowly, so that by four weeks after the beginning of treatment the average fasting concentration was 8 micrograms per liter. Individual curves frequently differed from the average pattern.

The average plasma concentrations at two and four hours after the administration of 0.1 Gm. of quinacrine hydrochloride with each meal varied from 2.3 to 12 and 6.8 to 11.3 micrograms per liter above the corresponding fasting levels.

During the period of suppressive treatment in the patients followed through the sixtieth day from the start of treatment (2.8 Gm.), the levels dropped slowly to an average concentration of 23 micrograms per liter.

The range of the daily average fasting quinacrine hydrochloride concentrations on the 3.2 Gm. treatment from the second through the eighth day was 36 to 63 micrograms per liter. When treatment was discontinued, the levels fell slowly, so that by the eighteenth day after treatment was started the average concentration was approximately that attained by the standard 2.8 Gm. group (15 micrograms per liter).

When a fasting plasma quinacrine concentration of approximately 45 micrograms per liter was attained within twenty-four hours and maintained, the symptoms of the attacks were usually abolished within seventy-two to ninety-six hours after the start of treatment.

Progress in Internal Medicine

BLOOD

A REVIEW OF THE RECENT LITERATURE

FRANK H. BETHELL, M.D.; CYRUS C. STURGIS, M.D.; R. WAYNE RUNDLES, PH.D., M.D., AND MURIEL C. MEYERS, M.D.
ANN ARBOR, MICH.

(Continued from Page 254)

IRON METABOLISM AND EXPERIMENTAL ANEMIA

Powell [51] determined the serum iron values for a series of 70 normal persons and for patients with various types of anemia. Mean values of 143 micrograms per hundred cubic centimeters for men and 117 micrograms for women were obtained. Evidence was obtained showing that the serum iron level falls during the menstrual period in normal women. The serum iron level was generally low in patients with hypochromic anemias, but after the beginning of treatment values as high as 400 micrograms per hundred cubic centimeters were found in many cases. In 2 patients a satisfactory hemoglobin response to iron therapy was obtained only after simultaneous administration of ascorbic acid. The author is inclined to attribute the benefit derived from the use of ascorbic acid to the reduction of ferric iron and its maintenance in the ferrous state during its passage into the upper part of the small intestine. One of the patients studied had hypochromic anemia refractory to iron therapy even when ascorbic acid was added, and the low serum iron values persisted through the period of treatment. The cause of this failure to absorb iron was not determined. The lowest value for serum iron, 15 micrograms per hundred cubic centimeters, was obtained for a patient with scurvy. High serum iron levels were found in patients with aplastic anemia and pernicious anemia. After the commencement of liver therapy for patients with the latter disease, levels as low as 30 micrograms were found. The values for serum iron in patients with hemolytic anemia were not significantly raised, and it was thought that in these circumstances breakdown and synthesis of hemoglobin were proceeding at approximately equivalent rates.

Moore and his collaborators,[52] using the radioactive isotope, reinvestigated the absorption of ferrous and ferric iron by human subjects and by dogs. Among physicians and clinical investigators there is agreement that patients with hypochromic anemia require from four to six times the quantity of iron in the form of a soluble ferric salt as that contained in a soluble ferrous preparation which must be given to produce a comparable therapeutic effect. Studies on the absorption of iron in human beings likewise have regularly shown that the bivalent form of the metal is more completely assimilated than is the trivalent. In investigations on animals less consistent results have been obtained, and some investigators have concluded that there is no difference in the degrees to which ferrous and ferric salts are absorbed. Six normal dogs were given 1 mg. per kilogram of body weight of either ferric or ferrous radioactive iron, the latter being reduced from the trivalent state by ascorbic acid. The appearance of the labeled element as newly synthesized hemoglobin in the peripheral blood was determined as a measure of the amount absorbed. In about half of the experiments the two forms of the iron were absorbed equally well, while in the remainder the ferrous form was assimilated better.

Similar doses of radioactive iron were given to 4 anemic dogs whose iron reserves had been depleted by repeated bleeding. A greater proportion of the total iron was absorbed by these animals than by those in the normal group, and there was approximately equal assimilation of the two valence forms.

Ferrous and ferric iron in test doses of 1, 2 and 4 mg. per kilogram of body weight were given to 8 different men with normal blood values. In each case more ferrous than ferric

Footnotes 49 and 50 have been renumbered and appear elsewhere in this review.

51. Powell, J. F.: Serum-Iron in Health and Disease, Quart. J. Med. **13**:19, 1944.

52. Moore, C. V.; Dubach, R.; Minnich, V., and Roberts, H. K.: Absorption of Ferrous and Ferric Radioactive Iron by Human Subjects and by Dogs, J. Clin. Investigation **23**:755, 1944.

iron was absorbed. The ratio of the amount of the bivalent form assimilated to that of the trivalent form assimilated varied from 1½ to 10. Similar studies were made on 5 patients with severe hypochromic microcytic anemia. The bivalent form was absorbed from two to fifteen times as well as the ferric form. The percentage assimilated was greater with the small doses than with the large, and the iron-deficient subjects tended to absorb a greater proportion of the iron than did the men with normal blood values.

Boyd, Hitsman and Perry [53] investigated the storage and elimination of iron compounds injected intramuscularly into albino rats. Of the iron injected none was lost or excreted in the feces, and only 1 to 2 per cent was excreted in the urine. About two thirds of the injected iron was stored in the liver, and smaller amounts were deposited in the walls of the intestines, the bones, the muscles and elsewhere.

Hahn and his collaborators [54] studied the absorption of red blood cells from the peritoneal cavity of dogs. Red blood cells labeled by radioactive iron in their hemoglobin were injected into the peritoneal cavity of normal dogs. Between 20 per cent and 100 per cent of the cells were found to be circulating in the blood within twenty-four hours. Anemic dogs absorb red cells in this manner a little less readily, and dogs depleted of both hemoglobin and protein absorb them even less readily. Although transit of the red cells from the peritoneal cavity to the circulation is surprisingly rapid, the rate varies widely in different dogs and in the same dog at different times.

An attempt to mobilize the iron in a patient with hemochromatosis by the administration of various chemicals was made by Fishback.[55] Over an experimental period of one week each the patient was given sodium thiosulfate, ammonium chloride and sodium bicarbonate by mouth and sodium thiosulfate intravenously while a careful chemical check of the iron balance was maintained. None of the compounds administered was found to have a definitely significant effect in mobilizing iron or in producing a negative iron balance.

Lucia and Marasse [56] reviewed at length the rather large volume of work done in evaluating the effect of the central nervous system on hemopoiesis. The authors concluded that a relation between the hypothalamus and erythropoiesis or between localized lesions or other diseases of the central nervous system and erythropoiesis has not been convincingly demonstrated.

Brown, Hayward, Powell and Witts [57] studied the rate of destruction of transfused erythrocytes by the Ashby method of differential agglutination. In 6 cases of idiopathic hypochromic anemia a nearly constant linear curve of destruction occurred. The transfused erythrocytes survived on the average fifty days. In 3 cases in which excessive hemolysis was apparent clinically, 1 that of a patient with a metastatic neoplasm and the other 2 of patients with acquired hemolytic anemias, the rate of hemolysis of the transfused red blood cells was nearly twice as rapid. The plotted values in the latter cases were curvilinear, with the most rapid hemolysis occurring during the first week. In 7 patients with anemia due to infection and in 12 other patients, single patients with such conditions as thyrotoxicosis, chronic nephritis and pernicious anemia under treatment with liver extract, variably accelerated rates of destruction of erythrocytes were found. The plotted curves were both linear and curvilinear in type. In most patients with acute anemia an unduly rapid rate of red cell destruction was found, while in patients with chronic anemia the rate of destruction was normal.

Higgins [58] designed a number of animal experiments using young male rats to test the value of vitamin B_c (folic acid) in correcting or preventing the anemia resulting from the administration of promin (sodium p, p′-diaminodiphenylsulfone-N,N′didextrose sulfonate) and promizole (4,2′-diaminodiphenyl-5′-thiazole-sulfone). The rats were fed a diet high in purified carbohydrate supplemented with thiamine, riboflavin, pyridoxine, calcium pantothenate, nicotinic acid and choline chloride. The addition of either of the two sulfone compounds to this diet had been shown to induce a hypochromic,

53. Boyd, E. M.; Hitsman, J. S., and Perry, W. F.: The Storage and Elimination of Iron Compounds Injected Intramuscularly into Albino Rats, Rev. canad. de biol. **3**:294, 1944.

54. Hahn, P. F., and others: Peritoneal Absorption: Red Cells Labeled by Radio-Iron Hemoglobin Move Promptly from Peritoneal Cavity into the Circulation, J. Exper. Med. **80**:77, 1944.

55. Fishback, H. R.: On the Mobilization of Iron in Hemochromatosis with Administration of Various Chemicals, J. Lab. & Clin. Med. **29**:742, 1944.

56. Lucia, S. P., and Marasse, H. F.: The Central Nervous System and Hematopoiesis, J. Nerv. & Ment. Dis. **99**:734, 1944.

57. Brown, G. M.; Hayward, O. C.; Powell, E. O., and Witts, L. J.: Destruction of Transfused Erythrocytes in Anaemia, J. Path. & Bact. **56**:81, 1944.

58. Higgins, G. M.: The Influence of Vitamin B_c Concentrate (Folic Acid) on Experimental Anemia in the Rat, Proc. Staff Meet., Mayo Clin. **19**:329, 1944.

although macrocytic, anemia in the rats. In 1 experiment, 80 micrograms of vitamin B_c per day was given orally to a group of rats receiving 50 mg. of promin daily. In another experiment, the anemia was first induced by giving promin in the same amount daily for two weeks. Vitamin B_c was then given orally to part of this group of animals for two weeks, while the remaining animals continued to receive promin but not the vitamin. A third experiment was conducted after the pattern of the second, with the anemia being first induced by giving promizole, 25 mg. per day. The author's findings appeared to indicate that under the experimental conditions set up vitamin B_c in daily amounts of 80 micrograms orally exerted a pronounced antianemic effect on the animals in which the anemia had been previously induced.

Michaud, Maass, Ruegamer and Elvehjem [59] studied regeneration of hemoglobin in dogs maintained on a purified sucrose-casein-synthetic vitamin ration to which succinylsulfathiazole in amounts varying from 0.5 to 4 per cent had been added. They attempted in this way to modify the intestinal flora and to cause a deficiency of factors possibly synthesized in the digestive tract. It had been found that dietary levels as low as 0.5 per cent of succinylsulfathiazole were injurious to rats under similar conditions. The hemoglobin content per hundred cubic centimeters of circulating blood, total hemoglobin, number of red blood cells, mean cell volume, food consumption, growth increase and the general state of health were the factors taken into consideration. In dogs they could find no difference between the animals receiving the drug and those serving as controls.

NUTRITION AND ANEMIA

Some of the fundamental relations between the plasma proteins, cell proteins and the protein constituents of hemoglobin are reviewed by Whipple and Madden.[60] In recent years a large number of observations have been made in their laboratory to show that a dynamic equilibrium exists between plasma protein and cell protein which enables protein to flow in either direction, depending on the conditions of the moment. All the nitrogen requirements of dogs, for instance, can be supplied by dog plasma given by vein. With large injections of plasma, hyperproteinemia did not develop and the albumin-globulin ratio was not altered. Other experiments showed that protein stored as cell protein was available for the manufacture of hemoglobin and plasma protein in dogs rendered hypoproteinemic. Some observations indicate that a plasma-depleted dog on a liberal protein diet can produce more new plasma protein, 70 Gm. or more per week, than a standardized anemic dog can produce hemoglobin, 70 Gm. or less per week. The body is able to make new hemoglobin and plasma protein out of materials contained in digests made from serum. Dogs are able to manufacture abundant plasma protein derived at least in part from dog or sheep hemoglobin when this is injected intraperitoneally. Casein digests administered in this way are also effective. Twenty-five to 40 Gm. of new hemoglobin and plasma protein may be produced from 100 Gm. of digest.

Cartwright, Wintrobe and Humphreys [61] report further studies on the anemia in swine due to pyridoxine deficiency. Deficiency of this vitamin had been shown to result in nutritional microcytic anemia in both dogs and pigs, an anemia which did not respond to administration of iron or copper but which was relieved by synthetic pyridoxine. Convulsions, ataxia, degeneration of the peripheral nerves and presence of hemosiderin in the spleen, liver and bone marrow had occurred in the experimental animals. An elevation of the plasma iron was found in animals deficient in pyridoxine, and a green pigment–producing substance was present in the urine which was found to be xanthurenic acid, a product derived from tryptophan metabolism.

The authors studied the effects of pyridoxine deficiency on the blood of swine in comparison to those of iron deficiency and of phenylhydrazine hemolytic anemia with inanition controls. The major conclusions were as follows: An increased rate of hemolysis does not occur in pyridoxine deficiency. By restricting the dietary intake of iron in pyridoxine-deficient animals, hemosiderosis could be prevented and the serum iron maintained at the low level seen in iron deficiency. The ataxia, convulsions, neurologic lesions and fatty liver characteristic of animals with pyridoxine deficiency were not altered by limiting the iron intake to produce a combined deficiency of both iron and pyridoxine. The

59. Michaud, L.; Maass, A. R.; Ruegamer, W. R., and Elvehjem, C. A.: Hemoglobin Regeneration in Dogs Receiving a Purified Ration Plus Succinylsulfathiazole, Proc. Soc. Exper. Biol. & Med. **56**:148, 1944.

60. Whipple, G. H., and Madden, S. C.: Hemoglobin, Plasma Protein and Cell Protein: Their Interchange and Construction in Emergencies, Medicine **23**:215, 1944.

61. Cartwright, G. E.; Wintrobe, M. M., and Humphreys, S.: Studies on Anemia in Swine Due to Pyridoxine Deficiency, Together with Data on Phenylhydrazine Anemia, J. Biol. Chem. **153**:171, 1944.

urinary excretion of iron in pyridoxine-deficient animals was not altered and remained insignificant. The increased amount of iron found in the blood serum was found to exist in the trivalent state. The administration of chlorophyll, purified liver extract, tryptophan, corn oil, crude hemoglobin, hemin and iron ascorbate were all ineffective in stimulating blood formation in this deficiency state. The authors regard the ferremia and hemosiderosis as being due to the continued absorption or decreased excretion of iron at a time when its utilization for hemoglobin formation is at a minimum and when the body tissues are abundantly supplied with iron.

Wintrobe, Follis, Alcayaga, Paulson and Humphreys [62] studied the effects of pantothenic acid deficiency in swine by maintaining the animals on a purified diet supplemented by all the known crystalline B vitamins other than pantothenic acid. With such a diet young pigs manifested signs of deficiency much sooner than did similar pigs receiving calcium pantothenate but lacking any one of the following: thiamine, riboflavine, nicotinic acid, pyridoxine or choline. Growth was quickly inhibited and diarrhea was severe and was accompanied with extensive pathologic changes in the colon. Sudden collapse occurred and ended fatally. Less pronounced effects of pantothenic acid deficiency in pigs were loss of hair, reddening of the skin, excessive nasal secretion, changes in the tongue and degenerative changes in the nervous system associated with the development of noticeable abnormalities in gait.

A severe anemia such as that which occurs in pyridoxine deficiency did not develop in the pantothenic acid–deficient pigs. A moderate normocytic anemia was noted in 13 out of 18 pigs. In 7 the volume of packed red cells ranged between 35 and 40 cc. per hundred cubic centimeters of blood, and in the remainder the volume of packed red cells ranged between 24 and 33 cc. (the normal range in pigs being 41.5 to 51.5 cc.). Two animals were treated with calcium pantothenate, and the anemia became less pronounced.

Maass and his collaborators [63] studied the relation of thiamine to blood regeneration in growing and in adult dogs. The animals were maintained on a purified diet supplemented with all the crystalline vitamin B fractions except thiamine and rendered anemic by repeated phlebotomy. No specific effect of the thiamine deficiency on the capacity for blood regeneration was found.

Bass [28] reports on 2 infants with nutritional anemias. In a 6 month old infant, weakness and pallor developed after an infection of the upper respiratory tract. The hemoglobin content of the blood was found to be 17 per cent and the red cell count 920,000. The infant was admitted to the hospital and given blood transfusions, iron and large doses of vitamins. There was, however, a progressive fall in the blood level during an observation period of four weeks. Injections of liver extract were then started, and the infant made a complete and uneventful recovery.

Another infant, 5 months old, had become severely allergic to cow's milk early in life and was given a goat's milk formula with vitamin B and C supplements. Anemia, with a hemoglobin content of 36 per cent and a red blood cell count of 1,630,000, was present at the age of 2½ months. This was treated by several small transfusions and by the addition of an iron preparation to the diet. The feeding problem and the severe eczema became worse, and the infant's condition became critical. The child was given a formula containing the amino acids derived from hydrolyzed casein with vitamin and iron supplements as before. In less than a month the cutaneous lesions had cleared and the blood levels were being maintained satisfactorily. It was the author's opinion that the hematologic improvement resulted from the more adequate protein nutrition provided by the amino acids.

Davidson and Donaldson [64] found mild degrees of anemia relatively common among English school children and investigated its response to treatment with iron and ascorbic acid. They found that the anemia responded well to the administration of as little as 15 grains (0.97 Gm.) of ferrous sulfate a week. A supplement of 25 mg. of ascorbic acid daily in addition had no effect in raising the hemoglobin level.

Pijoan and Elkin [65] investigated the cause of anemia among Shoshone Indian infants fed for prolonged periods on an exclusive milk diet. It was the tribal custom to give the infants nothing but breast milk until they were 1 or 2

62. Wintrobe, M. M.; Follis, R. H.; Alcayaga, R.; Paulson, M., and Humphreys, S.: Pantothenic Acid Deficiency in Swine, with Particular Reference to the Effects on Growth and on the Alimentary Tract, Bull. Johns Hopkins Hosp. **73**:313, 1943.

63. Maass, A. R.; Michaud, L., and others: The Relation of Thiamine to Blood Regeneration, Arch. Biochem. **4**:105, 1944.

64. Davidson, L. S. P., and Donaldson, G. M. M.: Treatment of Anemia in School-Children with Iron and Ascorbic Acid, Brit. M. J. **1**:76, 1944.

65. Pijoan, M., and Elkin, C. A.: Secondary Anemia Due to Prolonged and Exclusive Milk Feeding Among Shoshone Indian Infants, J. Nutrition **27**:67, 1944.

years of age and could eat the unmodified adult food. The breast milk from nursing mothers was found to contain but 1.7 to 1.8 mg. of iron per liter, an amount low enough to account for the prevailing anemia due to iron deficiency.

Civeira Otermin [66] studied the blood changes, particularly regarding the cellular elements and bone marrow, in 40 cases of chronic starvation with edema. In these cases the number of platelets in the peripheral blood was found to be decreased, the counts averaging 187,000. The average white blood cell count was 6,000, with a relative and absolute lymphocytosis and neutropenia. The studies of the bone marrow showed an increase in the number of megaloblasts with the erythrocyte-granulocyte ratio disturbed by an increase in the granulocytic elements. A certain degree of arrest of maturation in both series resulting in an increase in the less differentiated elements was observed. There was no alteration in the megakaryocytes.

According to Weder,[67] the Plummer-Vinson syndrome has been diagnosed very rarely in Switzerland, possibly because the syndrome has not been recognized. He reported on 4 women with the syndrome, whose ages ranged from 31 to 51 years. In the majority of the patients there were soreness and burning of the mouth and tongue, painful recurrent fissures at the angles of the mouth, difficulty in swallowing solid and even liquid foods and symptoms of anemia. All the patients had smooth tongues with atrophic papillae, perlèche and severe hypochromic anemia, with the hemoglobin reduced to 41 to 58 per cent of normal. In treatment the patients were given ferrous iron and a vitamin B preparation of high riboflavin content. There was prompt and permanent relief of their symptoms.

ANEMIA ASSOCIATED WITH PREGNANCY

In reporting a hematologic study of 48 cases of pregnancy and reviewing the literature on the anemias of pregnancy, Elliott [68] comments that most of the investigation in this field has been of two varieties: either a mass survey of a large series of cases or a detailed study of individual cases. His investigation was of the latter type. He selected for study 48 patients from an antepartum and obstetric clinic. Most of the patients were in poor economic circumstances, living in homes of semislum standard. Their diets were mainly carbohydrate. No patient was included if toxemia of pregnancy or other known organic disease was present.

Twenty of the patients were selected as "normals" on the basis of having a hemoglobin content of 80 per cent (Haldane) or over. The color indexes in this group, with 1 exception, fell between 0.8 and 1 and the mean corpuscular volume, also, with the 1 exception, between 73 and 88 cubic microns. The mean cell diameter showed an increased variability, indicating that an abnormal degree of anisocytosis was present in this group. The composite curve of the tests of erythrocyte fragility in hypotonic solutions of sodium chloride showed a slightly increased ease of hemolysis. The reticulocyte counts and plasma bilirubin values were not abnormal.

In the 28 pregnant patients whose values for hemoglobin were below 80 per cent, the value ranged from 32 to to 76 per cent and the red blood cell counts from 3,270,000 to 5,000,000 per cubic millimeter. The mean corpuscular volume ranged between 58 and 98 cubic microns. The standard deviation of the red cell diameters and the variability per cent were within normal limits in some persons of the anemic group, although the degree of anisocytosis was a striking feature in many of the films and was associated with considerable variation in staining density. Three patients had a median corpuscular fragility greater than the calculated normal and 21 greater than the observed normal. In the majority of the patients with anemia associated with pregnancy there was a distinct shift toward decreased fragility after parturition as compared with the antepartum reading. This change was observed as early as the fifth day after delivery and for 1 patient was delayed at least nine days. In only 2 patients with anemia was the reticulocyte count raised and then only to a maximum figure of 4 per cent. The plasma bilirubin value fell within normal limits.

The majority of the pregnant women with anemia responded to treatment in the expected manner. Eleven patients were given iron, and only 1, who had an initial color index of 1, failed to respond to both iron and an effective liver extract. Spontaneous improvement after delivery occurred. Four patients in all required blood transfusions.

The increase in median corpuscular fragility differentiates this anemia, irrespective of its other characteristics, from the majority of other anemias. There is as yet no explanation for this change. The abnormal degree of aniso-

66. Civeira Otermín, F.: Plaquetas, leucocitos y mielograma en enfermos de inedia crónica (enfermedad del edema), Medicina, Madrid (pt. II) **11**:194, 1943.

67. Weder, A.: Das Plummer-Vinsonsche Syndrom, Schweiz. med. Wchnschr. **73**:1354, 1943.

68. Elliott, G. A.: The Anaemias of Pregnancy: A Report on the Haematological Study of Forty-Eight Cases of Pregnancy with a Review of the Literature, J. Obst. & Gynaec. Brit. Emp. **51**:198, 1944.

cytosis occurring in these patients has been stressed previously as an important sign of abnormal hemopoiesis and is perhaps more significant than the hemoglobin level.

The occurrence of some degree of hydremia during pregnancy makes it possible for a hemoglobin content as low as 65 per cent (Haldane) and a red cell count as low as 3,200,000 to be considered physiologic provided certain other conditions are satisfied. The color index must be near unity, the mean cell diameter within normal limits, the values expressing the normal degree of anisocytosis and the mean corpuscular volume within the normal limits for pregnancy.

Several factors may account for the iron deficiency anemia of pregnancy: preexisting anemia, decreased secretion of hydrochloric acid in the stomach during pregnancy, poor social conditions and poor diet, changes in iron metabolism due to pregnancy and the number of previous pregnancies. The number of cases of megalocytic anemia of pregnancy reported in recent years has decreased, and further study is required to elucidate their causes. In the last ten years, not one example of acute hemolytic anemia of pregnancy has been reported. There still remain, however, a group of anemias occurring in pregnancy which it is difficult to fit into any known classification.

Piney [69] presents a short account of the blood disorders of most frequent occurrence during pregnancies, with an unusually good discussion of the accumulated information regarding the macrocytic anemias of pregnancy.

Wiener [70] offers a short comment regarding the macrocytic anemia of pregnancy, which has impressed some observers as resembling a hemolytic anemia. It is well known that the blood of every person contains, in addition to the agglutinogens characterizing the individual, one or more agglutinogens shared by all members of the species. Ordinarily a person does not become immunized or sensitized against antigens contained in his own body, but under certain conditions this principle of "horror autotoxicus" of Ehrlich breaks down. For example, when the sensitized Rh-negative patient is given Rh-positive blood, he breaks down the blood cells. Then the body in disposing of the broken-down cells may occasionally become sensitized to the common species of antigens contained in the stroma. The resulting antibodies have the properties of reacting not only with the blood of other individuals of the species but also with the patient's own blood cells. The antibodies then act on the patient's own cells, and a hemolytic anemia results. The patient's own broken-down cells may sensitize him still further, and continued hemolysis may result.

By extending this principle of "autoimmunization" to the conditions of pregnancy, a rather attractive explanation of the macrocytic anemias of pregnancy can be suggested. If an Rh-negative mother becomes oversensitized to the fetus' Rh-positive cells, she may also form antibodies to one or more of the common species antigens, and an acute hemolytic anemia could result. This theory stands in agreement with the known clinical characteristics of the disease, and in 3 cases of anemia of this type observed by Wiener the women all proved to be Rh-negative.

HEMOLYTIC ANEMIA

Congenital and Acquired Types.—The general clinical features of the hemolytic anemias, their classification and recent developments regarding their pathogenesis are the subjects of a review by Davis.[71] While in theory a hemolytic anemia is one in which the anemic state results mainly from the process of blood destruction proceeding at an accelerated rate beyond the capacity for blood regeneration, a definition of general value in clinical diagnosis is difficult. Many aspects of the mechanism concerned with the disposal of red blood cells and hemoglobin in health as well as in disease are still obscure. Concerning the destruction of red cells there is little agreement regarding the mechanism beyond the fact that cells damaged by age, trauma or special agents are disposed of by the phagocytic cells of the reticuloendothelial system, predominantly within the spleen. It is still uncertain whether the spleen is the "slaughterhouse" of the red cells, but there is a general consensus that the spleen is certainly their "graveyard." Following the breakup of the red cell, there is phagocytosis of the debris and iron is split off from the hemoglobin. The iron-free compound is converted into bilirubin, which circulates in the plasma, probably in combination with serum protein. The production of this hemobilirubin will obviously be increased under conditions of excessive hemolysis, and there may be increased output of fecal urobilinogen. The determination of the fecal urobilinogen is one of the most important recent advances in the study of the hemolytic anemias.

69. Piney, A.: Blood Disorders in Relation to Pregnancy, Post-Grad. M. J. **20**:170, 1944.

70. Wiener, A. S.: Macrocytic Anemia of Pregnancy, J. A. M. A. **125**:990 (Aug. 5) 1944.

71. Davis, L. J.: Haemolytic Anaemias, Edinburgh M. J. **50**:589, 1943.

Under certain pathologic conditions, hemolysis may be predominantly or exclusively intravascular, resulting in the liberation of free hemoglobin in the general circulation. The reticuloendothelial cells are capable of dealing with only a small amount of free hemoglobin, and when this limit is exceeded the excess breaks down into hematin and globin. Hematin combines with serum albumin to form methemalbumin, a compound which, because of its large molecule, never escapes into the urine. Its demonstration in the plasma by spectroscopic tests is a conclusive indication of intravascular hemolysis. The renal threshold for free hemoglobin is about 150 mg. per hundred cubic centimeters. Once hemoglobinuria is established, however, it may continue in the presence of a plasma hemoglobin concentration as low as 30 to 50 mg. per hundred cubic centimeters. Thus, hemoglobinuria is another indicator of intravascular hemolysis. While the finding of a sustained high reticulocyte count, nucleated red blood cells, anisocytosis and so forth in the peripheral blood and hyperactive erythropoiesis in the bone marrow are suggestive of excessive hemolysis, unequivocal evidence of such an occurrence may be obtained by demonstrating an increased excretion of fecal urobilinogen, methemalbuminemia or hemoglobinuria.

In familial acholuric jaundice and in sickle cell anemia conspicuous erythrocyte abnormalities are associated with the hemolytic tendency, but the details of the mechanism in each disease are unknown. Nocturnal hemoglobinuria is a form of chronic hemolytic anemia often of considerable severity and displaying a fluctuating course. That intravascular hemolysis occurs has been demonstrated by the finding of increased plasma hemoglobin and methemalbumin. The hemolytic phenomena result from an abnormality of the red blood cells which renders them peculiarly liable to lysis by some agent normally present in the plasma, probably a complement. This susceptibility is aggravated by an increased hydrogen ion concentration, such as occurs during sleep consequent on reduced pulmonary ventilation. Davis had the opportunity to study a patient with this disorder, who finally died after an exacerbation of the disease. The effect of p_H in producing hemolysis could be demonstrated by placing a specimen of the patient's heparinized blood in a tube layered over with paraffin to prevent the loss of oxygen. Hemolysis was evident after a few hours, while control tubes containing normal blood or the patient's blood without paraffin showed no hemolysis.

Hemolytic anemias may result from circulating hemolysins, such as may follow the transfusion of incompatible blood, sensitization by the Rh antibody, activation of a specific hemolysis in some syphilitic persons by exposure to cold, severe burns, bacterial infections (especially those due to anaerobic organisms) or snake venom. Parasitic infections such as malaria, which is by far the commonest cause of hemolytic anemia, poisons such as lead, phenylhydrazine and phenothiazine and sensitivity to drugs or plants are other etiologic factors which must be considered in diagnosis.

The so-called acquired hemolytic anemias, acute and chronic anemias of unknown cause, comprise a heterogenous group differing widely in their hematologic and clinical aspects. The prognosis is grave and the mortality rate high. In some cases transfusion may be beneficial or curative and in others of little benefit and productive of hazardous reactions, but this measure should always be tried. The results of splenectomy have varied in different series, and no final evaluation is possible.

Davis [72] reports 4 cases of symptomatic or acquired hemolytic anemia with increased urobilinuria, hyperbilirubinemia and reticulocyte counts from 22 to 66 per cent in persons with other primary diseases. Most of them had a personal or family history of jaundice or other manifestation of familial hemolytic icterus. In the first case, a 72 year old man complained of pain in the back and increasing fatigue and weakness and showed pallor, jaundice and hepatomegaly. A severe macrocytic anemia was present, and the reticulocyte count was persistently elevated. Despite treatment he died in a few weeks' time, and necropsy showed that a carcinoma of the pancreas was present, with metastases to the liver, one kidney and the retroperitoneal, mediastinal and cervical lymph nodes. The second case was that of a 19 year old youth who had received roentgen ray therapy three years earlier for a cervical swelling thought to be lymphadenoma. Progressive weakness and pallor developed, and on examination light jaundice, slightly enlarged inguinal nodes and a palpable spleen were found. A normochromic but slightly macrocytic anemia was present, with reticulocytosis. The illness terminated fatally in a few weeks. Permission for an autopsy was not obtained. The third case was that of a 50 year old man in whom a severe hemolytic anemia developed a few weeks after a generalized enlargement of the lymph nodes and splenomegaly had appeared. He had been given a course of

72. Davis, L. J.: Symptomatic Haemolytic Anaemia: A Report of Four Cases, Edinburgh M. J. **51**:70, 1944.

sulfapyridine without apparent effect. He improved with roentgen therapy and blood transfusions, and during the remaining three years of his life there were no hematologic abnormalities. The diagnosis at autopsy was advanced Hodgkin's disease. The fourth case was that of a 41 year old woman who after being ill for a year with anemia was found to have subleukemic myeloid leukemia. Excessive red blood cell destruction and increased blood regeneration were constantly present during the last week of her illness. Autopsy confirmed the diagnosis of myeloid leukemia. The cause of the hemolysis in these cases remained obscure. There was no evidence of pronounced erythrophagocytosis. It was thought that the tumor growths might have exerted an injurious effect on the circulating erythrocytes, promoting their destruction, or that the activity of the reticuloendothelial cells might have been abnormally stimulated.

A confusing case of symptomatic hemolytic anemia is reported by Feldman and Yarvis.[73] An 18 year old youth complained of bleeding from the gums and passing dark-colored urine. Examination showed that he was jaundiced and that his spleen was greatly enlarged. The hemoglobin content was but 60 per cent of normal, the red blood cells 2,750,000 and the white blood cells 25,000. There were microspherocytosis and reticulocytosis, and the erythrocyte fragility was increased. The differential count included 30 per cent young lymphocytes and 10 per cent abnormal lymphocytes, which were considered to be characteristic of those of infectious mononucleosis. The heterophile agglutination test was repeatedly positive up to a dilution of 1:1,024. Two other hematologists reviewed the findings and agreed in the diagnosis of infectious mononucleosis and hemolytic icterus.

The patient continued to get worse, had many hemorrhagic episodes and required repeated blood transfusions. Finally splenectomy was advised, and a spleen weighing 2,000 Gm. was removed. The patient improved temporarily but then failed again. At this time the blood picture was typical of lymphatic leukemia, with the white cell count rising to 230,000.

Neumann[74] reports a somewhat similar case, that of a 51 year old man, who became jaundiced and in whom a hemolytic anemia was found. Biopsy of an enlarged lymph node was performed, and a diagnosis of lymphogenous leukemia made.

Concerning the pathogenesis of familial hemolytic icterus, Willenegger[75] discusses the two factors of fundamental importance: (1) the primary, inherited or constitutional anomaly of the red blood cells manifested by their spherocytic forms and diminished osmotic resistance to hypotonic sodium chloride solutions and to lysolecithin and (2) an abnormal activity of the spleen in the destruction of erythrocytes. The constancy of splenomegaly in this disease and the therapeutic benefit of splenectomy illustrate the importance of the splenic factor in the pathogenesis of the disorder, even though splenectomy as a rule does not alter the spherocytosis or increased red blood cell fragility. Some authors have emphasized the function of the spleen in storing erythrocytes in this connection, especially since the spherocytic form and diminished osmotic resistance of the red blood cells occur during their sojourn in this organ. An experiment was planned to evaluate these factors in a patient who was undergoing splenectomy. Seventeen hours before operation 300 cc. of fresh type O blood was given to the patient, whose blood was type A. By means of an anti-A hemolysin the donor's and the recipient's erythrocytes in the patient's circulating blood and in the splenic sinuses were later separated for cell counts and morphologic observations.

The majority of the transfused erythrocytes were found to be sequestered in the spleen by the time operation was performed. Those that remained in the circulating blood disappeared somewhat more quickly than would normally be anticipated after transfusion. The transfused erythrocytes recovered from the splenic sinuses seventeen hours after they were injected had already become spheric and even crenated. The donor's cells that remained in the circulating blood showed little or no morphologic change. Blood corpuscles suspended in plasma obtained from the spleen of the person with familial hemolytic icterus showed no morphologic change different from that of those suspended in normal plasma. The author interprets his observations as indicating that the spleen plays an active role in the pathogenesis of familial hemolytic icterus.

Wiedemann[76] discusses the relation of microspherocytosis to decreased osmotic resistance of

73. Feldman, F., and Yarvis, J. J.: Manifestations of Hemolytic Phenomena and Infectious Mononucleosis in a Case of Lymphatic Leukemia, New York State J. Med. **44**:1693, 1944.

74. Neumann, H.: Lymphadenom und hämolytischer Ikterus, Klin. Wchnschr. **20**:669, 1941.

75. Willenegger, H.: Zur Pathogenese des konstitutionellen hemolytischen Ikterus, Schweiz. med. Wchnschr. **74**:491, 1944.

76. Wiedemann, H. R.: Familiärer hämolytischer Ikterus und osmotische Hämolyse, Ztschr. f. Kinderh. **63**:501, 1942.

the erythrocytes in familial hemolytic icterus. In afflicted families other authors have found as many as 10 per cent of the known carriers of the disease to have normal results of fragility tests. He investigated the family of a 12 year old boy who had had intermittent jaundice from the age of 8 weeks. Both the liver and the spleen were enlarged. A moderately severe anemia was present, with 7 to 15 nucleated red blood cells per hundred white blood cells and a reticulocytosis of 24 to 32 per cent. The mean corpuscular volume was 107 cubic microns. There was no increase, however, in the level at which hemolysis began, although the point of complete hemolysis was definitely above that of the normal control. The mother of the patient and 3 of his sisters had either splenomegaly or jaundice, but there was no alteration in the hypotonic fragility of their cells except for a slightly higher level at which complete hemolysis occurred. The degree of spherocytosis was not pronounced in this family. The author regarded the decreased maximal resistance of the red blood cells as a significant point in diagnosis in atypical cases.

Ducci, Etcheverry, Nijamkin and Zanartu [77] report their observations on 2 brothers who had a form of familial icterus which they considered not to be of hemolytic origin. In 1 boy the features resembled those of familial hemolytic icterus, since there was an elevated blood bilirubin, a slight increase in the red blood cell fragility and an increased elimination of fecal urobilinogen. In the other patient icterus had been present for several years, but the blood values were normal; there was no splenomegaly, and tests of hepatic function, including that of bilirubin excretion, produced normal results. Other relatives were known to be icteric, but they were not available for study.

Horne [78] reviews the general aspects of familial hemolytic icterus of especial interest to surgeons concerned with splenectomy and with the treatment of complications of the biliary tract. When the diagnosis of familial icterus is established, splenectomy is ordinarily advisable at an early age. In competent hands, the operative mortality is low and permanent benefit results. In about 60 per cent of untreated patients with long-standing disease gallstones develop, and in every case a roentgen examination of the gallbladder should be made or the organ palpated for stone at the time of splenectomy. Perforation of the gallbladder in these cases and the formation of sinus tracts following cholecystotomy occur frequently. Two cases are reported in detail. In 1, a 16 year old girl had recurrent jaundice from early life, and biliary colic developed when she was 15 years old. A splenectomy and cholecystectomy were done in two stages, and the final clinical result was good. The second patient, a 17 year old youth, had had fairly good health in spite of recurrent jaundice and a chronic hemolytic anemia until biliary complications developed. He was operated on for cholecystitis, and the gallbladder, which contained stones, was drained. A cholecystotomy sinus persisted, with periodic passage of stones, and finally jaundice reappeared. The gallbladder was found to have ruptured, causing a bile peritonitis, and a fatal outcome resulted. Splenectomy with early cholecystectomy when gallstones are present is the treatment of choice for this disease.

Hobbs [79] reports 2 cases of march hemoglobinuria occurring in soldiers during training. One 20 year old soldier noted that he passed "bloody" urine after nearly every severe exertion for a period of seventeen days. A slight amount of pain in the region of the left flank accompanied the voiding of the abnormal urine. After the patient was confined to the hospital, several specimens of urine were examined without any abnormality being found and results of a urologic investigation were negative. He was then sent on a trial hike of 10 miles (sixteen kilometers) carrying a pack weighing 34 pounds (15.4 Kg.). "Bloody" urine was passed after he had hiked 5 miles (8 kilometers). A specimen of urine obtained on completion of the hike was the color of port wine; the reaction for albumin was 4 plus; the benzidine reaction was 4 plus, and 3 Gm. of hemoglobin were present per hundred cubic centimeters of urine. Only an occasional white blood cell was present in the urinary sediment. The hemoglobin content of the plasma rose from 6.2 mg. per hundred cubic centimeters before the hike to 170 mg. after the hike. The fragility of the red blood cells in hypotonic solutions of sodium chloride was unaltered. A second soldier, a 32 year old Negro, stated that he had passed "bloody" urine at least twelve times in five months, each time after strenuous exertion. A specimen of urine obtained after a hike was the color of port wine; the reaction for albumin was 2 plus and the benzidine reaction 4 plus. The urobilinogen test showed a

77. Ducci, H.; Etcheverry, R.; Nijamkin, A., and Zanartu, J.: Istericias familiares hemolítica y no hemolítica, Rev. méd. de Chile **71**:837, 1943.

78. Horne, E. O.: Congenital Hemolytic Icterus: Surgical Treatment of Complications with a Report of Two Cases, Am. J. Surg. **65**:56, 1944.

79. Hobbs, R. E.: March Hemoglobinuria: A Report of Two Cases, Am. J. Clin. Path. **14**:485, 1944.

reaction in a dilution of 1:20. The hemoglobin content of the plasma was 80 mg. per hundred cubic centimeters. The results of tests of red blood cell fragility with hypotonic sodium chloride and acid were normal.

The author determined the plasma hemoglobin level in 23 men just completing a speed march of 10 miles and found values ranging from 15 to 25 mg. per hundred cubic centimeters. In the diagnosis of march hemoglobinuria the significant and constant observations are hemoglobinemia and hemoglobinuria. Concerning its pathogenesis, the author favors the theory of a general intravascular hemolysis.

Makin [80] reports a case of march hemoglobinuria in a 21 year old infantryman which he observed. The soldier noted that he began to pass dark-colored urine after strenuous activity in training and in sports. A urinalysis done after rest showed nothing abnormal. After exercise, however, his urine became dark brownish red in color; the benzidine reaction became positive, and a heavy albuminuria was present, as well as granular casts in the sediment. Spectroscopic examination showed that the urine contained oxyhemoglobin. In four months' time the hemoglobinuria following exercise gradually decreased and finally disappeared. The soldier was subsequently returned to full army duty without ill effects.

Bryce [81] reports a similar case of march hemoglobinuria occurring in an 18 year old soldier. In this patient also aching pain in the groins accompanied the passage of dark urine. This occurred only after unusually strenous training exercises had been performed. A complete urologic examination was made and no anatomic abnormality found. Finally to establish the diagnosis urinalyses were made after heavy exercise. Under these conditions albumin, free hemoglobin and methemoglobin were present in the urine. The Donath-Landsteiner test failed to reveal paroxysmal hemoglobinuria. Experiments were conducted to determine the amount of work that could be tolerated without producing the hemoglobinuria. The soldier could tolerate working in a stooped position, as in vigorous digging, for at least two hours without symptoms, whereas walking for one to one and a half hours at a brisk pace brought on an attack.

In a discussion of chronic hemolytic anemia with nocturnal hemoglobinuria, Hegglin [82] states that no form of therapy yet available, including the administration of alkalis and splenectomy, modified the course of the disease materially. He recommended as a palliative measure the use of frequently administered small blood transfusions.

Losada, Etcheverry and Uauy [83] report the case of a 16 year old girl with paroxysmal hemoglobinuria. For five years during the seasons of cold weather she had recurrent episodes of chills, fever and passage of red urine, followed by pallor and icterus. She had had an unusual number of childhood diseases; and when she was 14 years old, enlargement of the cervical lymph nodes developed and examination revealed a tuberculous pleural effusion. The examination of the blood revealed an erythrocyte count of 1,500,000, a hemoglobin value of 50 per cent and a leukocyte count of 2,400. Reticulocytes were increased to 14 per cent; nucleated red blood cells were present, and the average cell volume was 145 cubic microns. The Kahn, Wassermann and Donath-Landsteiner reactions were strongly positive. While she was under treatment for the tuberculosis, there was progressive improvement in the hematologic status.

Hegglin and Maier [84] describe a "heat resistance" test as a specific test for the recognition of Marchiafava's anemia. This rare disease is characterized chiefly by nocturnal hemoglobinuria and the manifestations of a hemolytic anemia. The hemolysis is produced by the action of a hemolysin whose amboceptor is always present in the erythrocytes but which requires the presence of complement. The degree of hemolysis increases as the temperature of the blood rises and as the p_H of the blood falls.

The test is designed to determine the resistance of the red blood cells to increased temperature. It is performed by putting a test tube containing about 5 cc. of blood obtained by means of a syringe sterilized by dry air into the incubator for six to twenty-four hours at a temperature of 37 C. The degree of hemolysis can be observed with the naked eye. Although a quantitative determination of the hemoglobin liberated in the serum is for practical purposes unnecessary, it was done in a series of cases to

80. Makin, M.: A Case of March Haemoglobinuria, Brit. M. J. **1**:844, 1944.

81. Bryce, L. M.: March Haemoglobinuria: Description of the Features of This Condition, and Report of a Case, M. J. Australia **2**:49, 1944.

82. Hegglin, R.: Die chronische hämolytische Anämie mit nächtlicher Hämaglobinurie (Typus Marchiafava), Helvet. med. acta **10**:27, 1943.

83. Losada L., A.; Etcheverry, R., and Uauy A., N.: Hemoglobinuria paroxística esencial (a propósito de un caso clínico asociado a tuberculosis pleural), Rev. méd. de Chile **71**:1113, 1943.

84. Hegglin, R., and Maier, C.: The "Heat Resistance" of Erythrocytes: A Specific Test for the Recognition of Marchiafava's Anemia, Am. J. M. Sc. **207**: 624, 1944.

prove the specificity of the test. Two patients with Marchiafava's anemia had serum hemoglobin values of 570 and 675 mg. per hundred cubic centimeters after six hours' incubation and 592 and 1,375 mg. after twenty-four hours' incubation. The highest value in six other types of hemolytic anemia was 180 mg. per hundred cubic centimeters, found after twenty-four hours' incubation. In 50 normal persons, 7 patients with secondary anemia and 12 patients with pernicious anemia there was no increase in hemolysis. The possibility of increased acidity being produced by the heat and causing the hemolysis was not investigated.

Meier [85] reports 2 cases of acute hemolytic anemia of the Lederer-Brill type, which occurred in children. A 3 year old boy, who had previously been well, in a matter of two days' time became greatly fatigued, refused to eat and was constantly drowsy. Examination showed the child confined to bed, febrile, difficult to arouse, pale and jaundiced. Both the liver and the spleen were enlarged. The examination of the blood showed the hemoglobin content to be 20.3 per cent, the red cell count 575,000 and the white cell count 24,400. The reticulocytes were 40 per cent, and many nucleated red blood cells and spherocytes were present. The erythrocyte fragility in hypotonic sodium chloride solutions was slightly increased. Immature myeloid cells were present in the circulating blood. The Wassermann reaction was negative. Intramuscular injections of blood were given on two successive days, as well as *Néoton-Hépar,* iron therapy and symptomatic medication. The child began to improve in three days, was playing actively by the sixth day and later made a complete recovery. There was a transient eosinophilia rising to 16.5 per cent, but the blood picture finally became entirely normal. The parents were given physical examinations and examinations of the blood but no abnormalities were found. The child was still well when last seen sixteen months after the acute illness.

The second child was a 4 year old boy who had had good health until four or five weeks before his admission to the hospital, when fever, obstipation and cramping abdominal pain developed. Shortly afterward he became pale, listless and slightly jaundiced. His condition did not improve. Physical examination showed the boy to be poorly nourished and extremely pale and to have a yellowish complexion. The abdomen was distended, and neither the liver nor the spleen was palpable. Examination of the blood showed the hemoglobin content to be 16 per cent, the red cell count 1,100,000 and the white cell count 12,500, without unusual immaturity of the myeloid cells. The reticulocytes were increased to 15 per cent, and there were 6 nucleated red blood cells for every 100 white blood cells. There were no spherocytes. It was thought that rupture of the appendix with abscess formation had occurred and that the hemolytic anemia was secondary to this process. Two blood transfusions, sulfathiazole and *Néoton-Hépar* were used in therapy, and the boy recovered in two months.

The author discusses many of the other reported cases of acute hemolytic anemia in children and points out that a dramatic response to blood transfusion is by no means invariable.

Spira [86] reports the case of a 20 month old child in whom an acute hemolytic anemia developed fourteen days after a febrile illness, with a cutaneous rash and mild otitis media thought to be scarlet fever. Vomiting, high fever and the passage of dark red urine marked the onset of acute anemia. The child became extremely pale, icteric and finally moribund. The hemoglobin value was 16 per cent, and there was a leukocyte count of 34,000. The erythrocytes showed diminished resistance in hypotonic sodium chloride solutions. A remarkable improvement occurred with the first blood transfusion and subsequently a complete hematologic recovery occurred. An intracutaneous tuberculin reaction was found to be strongly positive, and four weeks after the onset of the anemia there was roentgen evidence of enlargement of the paratracheal lymph nodes. This began to recede after eight weeks of rest in bed, but the primary tuberculous process was kept under observation.

Videla and Bonell [87] report 2 cases of acute hemolytic anemia in adults complicating infectious processes. A 20 year old woman became acutely ill with nausea, diarrhea, abdominal cramps, fever and jaundice. Examination showed that pneumonia with pleuritis appeared to be the primary disease. Anemia, hemolytic in type, with erythroblastosis was present. A single blood transfusion was given, and the patient proceeded to make an uneventful recovery. A similar train of events was observed in a case of a 42 year old man suffering from an infection of the hand with lymphangitis.

85. Meier, K.: Die akute hämolytische Anämie vom Typ Lederer-Brill, Ann. pædiat. **162**:140, 1944.

86. Spira, M.: Zur Frage der akuten, hämolytischen, infektiösen Anämie, Typ Lederer im Kindesalter, Ztschr. f. Kinderh. **64**:187, 1943.

87. Videla Z., P., and Bonell, J.: Eritroblastosis reaccional; 2 casos, Rev. méd. de Chile **71**:1204, 1943.

An unusual case of acute hemolytic anemia in a man showing the phenomenon of autoagglutination and autohemolysis at room temperature was studied by Currie.[88] A 37 year old engineer had enjoyed excellent health until he suddenly and without warning felt faint while walking to work. He was forced to sit down, and on attempting to get up he collapsed and lost consciousness for a time. A few hours later progressively deepening jaundice appeared, along with hemoglobinuria. He was hospitalized two days later, when disorientation, confusion and fever had developed. Physical examination revealed extreme weakness, a thready pulse and a tender, palpable spleen. The urine contained hemoglobin and a large amount of hemoglobinous deposit but no bilirubin. Examination of the blood showed that hemoglobin content was 15 per cent, the red blood cell count 900,000, the leukocyte count 25,000 and the icteric index 34. In the blood film there were many normoblasts, some erythroblasts and immature granulocytes. There was obvious microspherocytosis.

The patient's serum was found to agglutinate his own corpuscles at room temperature. His serum agglutinated both group A and group B corpuscles but not normal group O corpuscles. When the blood was typed at body temperature, the patient's cells were found to contain neither A nor B agglutinogen. An autohemolysin was shown to be present in the patient's serum which required the presence of complement for its activity. The patient made an uneventful recovery, receiving 1 pint (0.47 liter) of whole citrated blood on the fourth day after his admission to the hospital, at which time he was apparently improving spontaneously. The phenomena of autoagglutination and autohemolysis disappeared during the convalescent period.

Foy, Gluckman and Kondi [89] report a case in which acute erythrocyte hemolysis and severe anemia followed the first administration of benzyl sulfanilamide. Icterus appeared after 12 Gm. of the drug had been given, and dark-colored urine was passed after 3 more Gm. had been given. Fever and tachycardia appeared, and when a blood transfusion was attempted a severe reaction developed. Oliguria, hypertension and nitrogen retention developed, and the patient died seven days after the initial reaction. At autopsy the kidneys were found to be deep purple, extremely congested and edematous. There was no mechanical obstruction present in the larger urinary passages, and in histologic sections there was no blockage of the renal tubules with precipitated products of hemoglobin. The authors discuss the blood pigment metabolism in persons with such conditions as blackwater fever, hemolytic jaundice and sulfonamide hemolysis. Oxyhemoglobin, methemalbumin and hemobilirubin are commonly found in all types of intravascular hemolyses. Intracorpuscular methemoglobin does not occur in blackwater fever, as far as is known. In the case reported, the intravascular hemolysis and rapid fall in the red cell count were accompanied with the presence of plasma oxyhemoglobin, methemalbumin and hemobilirubin as well as of intracorpuscular methemoglobin. Oxyhemoglobin and methemoglobin were found in the urine.

Cole [90] points out that blackwater fever may cause death in two different ways: (1) by fatal anemia due to erythrocyte hemolysis and (2) by renal damage leading to uremia. He presented the case of a young Indian who had a second attack of malaria, for which he took 5 grains (0.32 Gm.) of quinine daily. Four days after the onset of the chills and fever, he began to pass dark urine containing a brown deposit, a large amount of albumin and casts. He soon became jaundiced; the hemoglobin content fell to 70 per cent of normal, and the urine became scanty and the color of port wine. Four days later the blood urea level was 104 mg. per hundred cubic centimeters, and the urine, although only a few cubic centimeters per day was passed, was clear. A further rise in the blood urea nitrogen to 386 mg. per hundred cubic centimeters and generalized edema developed before the urinary output increased and recovery ensued. Four weeks after the onset of the illness, the urine had again become normal, and the urea clearance test showed no evidence of impaired renal function.

Strumia [91] discusses the hemolytic reactions which follow the transfusion of incompatible blood. When transfused blood is incompatible, the recipient's plasma agglutinates and hemolyzes the donor's cells. Clinical signs of a hemolytic reaction start during or shortly after the transfusion and commonly consist of a chill, followed by nausea, vomiting, pain in the lumbar region, a sense of constriction in the chest and fever.

88. Currie, J. P.: Acute Haemolytic Anaemia: Report of a Case Presenting Hitherto Unreported Features, Brit. M. J. **2**:8, 1944.

89. Foy, H.; Gluckman, J., and Kondi, A.: Pigment Metabolism and Renal Failure in Acute Sulphonamide Haemolysis Resembling Blackwater Fever, Tr. Roy. Soc. Trop. Med. & Hyg. **37**:303, 1944.

90. Cole, A. C. E.: A Case of Anuria in Blackwater Fever, East African M. J. **20**:381, 1943.

91. Strumia, M. M.: Post-Transfusion Hemolytic Reactions, Pennsylvania M. J. **47**:668, 1944.

Transient hemoglobinemia followed by oliguria is common. The oliguria may rapidly improve and the patient recover, or azotemia may result and recovery or death ensue after several days. A number of patients have died even though the urinary output has become normal or even greater than normal.

Whenever doubt exists that a hemolytic reaction has occurred, the first urine passed should be examined for albumin, hemoglobin, casts and red blood cells. The blood level of bilirubin should be determined immediately and after five and twelve hours, during which time the peak of bilirubinemia is generally reached.

Strumia does not subscribe to the theory that blockage of the renal tubules accounts for the renal damage associated with transfusion reactions or that alkalinization of the urine is of therapeutic usefulness. The outstanding pathologic lesions in such cases are extremely severe degenerative changes in the epithelium of convoluted tubules, with interstitial edema and vascular embolic phenomena with thrombosis. Since free hemoglobin per se is nontoxic, it appears likely that some factor in the "stroma" of the erythrocytes is toxic, perhaps the A, B or Rh agglutinogens. No therapy has been uniformly successful in treating the renal damage following transfusion of incompatible blood.

The prognosis is poorer for those patients having previous kidney damage or those with other serious illnesses. Most patients receiving 250 cc. of blood or less will recover from their first hemolytic transfusion reaction. Those receiving as much as 500 cc. or those who have had previous transfusion reactions will usually die.

Precautionary measures include meticulous typing and cross matching, with attention to the Rh factor and to the less common isoagglutinins in special cases. Slow transfusions are advised. Plasma should be used in emergencies rather than blood, even than blood from low titer group O donors with agglutinins not in excess of 1:60.

A serious posttransfusional reaction is reported by Introzzi and Bellotti.[92] A 30 year old woman was prepared for a cholecystectomy for biliary stone. At operation pericholecystitis with a greatly thickened gallbladder wall was found, and the removal of the gallbladder was accomplished with difficulty. A postoperative hemorrhage occurred, and 250 cc. of group O blood was given without apparent reaction. The following day restlessness, drowsiness and subicterus developed. The urinary output gradually fell until it was less than 250 cc. per day. Urinalysis gave positive reactions for occult blood, and a few red blood cells were present in the sediment. Lethargy persisted and edema and nitrogen retention were present for several days, but the patient eventually recovered.

Rh Factor.—Wiener, Sonn and Belkin[93] report further study of the heredity of the Rh blood types. Having available three varieties of anti-Rh agglutinins, they were able to demonstrate five kinds of agglutinogens which in combination determine eight types of human blood. Wiener has proposed a theory of six allelic genes to account for the existence of these variants of the Rh agglutinogen. The six gene theory postulates the existence of a series of allelic genes, Rh_1, Rh_2, Rh′, Rh″, Rh and rh, named after the serologic properties which they determine. The gene rh is recessive to the others and in homozygous persons gives rise to the Rh-negative type. The gene Rh′ determines an agglutinogen Rh′, which reacts with anti-Rh_1 but not with anti-Rh_2 or with anti-Rh agglutinins. Similarly the agglutinogen determined by the gene Rh″ reacts only with the anti-Rh_2 agglutinin, while agglutinogen Rh reacts only with the standard anti-Rh agglutinin, not with anti-Rh_1 or anti-Rh_2. The agglutinogens determined by the genes Rh_1 and Rh_2, on the other hand, are each characterized by their ability to react with two of the three primary agglutinins. For example, agglutinogen Rh_1 reacts with agglutinins anti-Rh_1 and anti-Rh but not with agglutinin anti-Rh_2. The capacity of bloods of types Rh_1 and Rh_2 each to react with two distinct agglutinins is inherited as a unit and is assumed to be due to the action of single genes.

With the six gene theory, twenty-one genotypes are theoretically possible. If the phenotype corresponding to each genotype is assumed to represent merely the combined effect of the two genes, then the twenty-one genotypes give rise to eight phenotypes, all but one of which has actually been encountered.

The authors investigated the Rh blood types in 97 families with 275 children and in 135 mother-child combinations. The results obtained were in complete agreement with the theory of six allelomorphic genes. With regard to linkage relations, the authors reported that the genes are transmitted independently of sex, the A-B-O genes and the M-N genes. When active and specific serums are generally available, many

92. Introzzi, A. S., and Bellotti, C.: Hemolisis posttransfusional con insuficiencia renal grave; plasmoterapia, Bol. y trab., Acad. argent. de cir. **28**:6, 1944.

93. Wiener, A. S.; Sonn, E. B., and Belkin, R. B.: Heredity of the Rh Blood Types, J. Exper. Med. **79**: 235, 1944.

clinical applications of these observations will be possible—for example, in transfusion therapy and in the classification of donors. The determination of the Rh type of the father of an erythroblastotic baby will often make possible a more definite statement as to the prognosis of future pregnancies. If the father belongs to type Rh_1-Rh_2, all the children must be Rh positive, half Rh_1 and half Rh_2 in the usual case; hence the prospect of the sensitized mother's bearing normal infants with this father is extremely remote. The medicolegal application of the Rh tests and their use in anthropologic studies and in studies of heredity are likewise practical.

Wiener [94] discusses in another report the role of the Rh subtypes in hemolytic transfusion reactions and in erythroblastosis. Analogous problems are presented by the two conditions. In hemolytic disease of the newborn, the Rh antigen is involved in about 90 per cent of the cases. The author encountered an infant with severe erythroblastosis whose mother was reported to be Rh positive. A more complete investigation showed that the father's type was Rh_1Rh_2 and the child's Rh_1Rh_2, while the mother's was actually Rh′. Thus isoimmunization of the Rh-positive mother by Rh-positive fetal blood of a different subtype was proved to be the mechanism responsible for the occurrence of erythroblastosis in the infant. The infant responded well to a blood transfusion of the mother's washed red cells suspended in the father's plasma. In such transfusions it is always safe to use the mother's washed red cells, while it is only generally safe to use Rh-negative blood. Another important corollary of these observations is that Rh-negative blood should be used in transfusions for the mother of an erythroblastic infant even though her blood is Rh positive in tests with the human Rh antiserum on hand. Another case of hemolytic disease of the newborn is described, in which the hemolysis was caused principally by Rh isoantibodies acquired by the infant through ingestion of the maternal milk.

Boorman, Dodd and Mollison [95] studied the incidence of hemolytic disease of the fetus in different families and the value of serologic tests in diagnosis and prognosis. The main features of the relation of the Rh antibody to the hemolytic disease of the newborn are well established, but much remains to be learned. Although the combination of an Rh-positive infant and an Rh-negative mother occurs in about 9.5 per cent of all pregnancies, the infant is affected with hemolytic disease in only a small proportion of the cases, for the incidence of hemolytic disease is only 1 in 400, or 0.25 per cent of all pregnancies. The mere demonstration that the mother is Rh negative and the infant Rh positive cannot by itself be regarded as satisfactory diagnostic evidence of hemolytic disease of the fetus. Moreover, the mother has been found to be Rh positive in a small proportion of cases in which the infant was affected with the disease.

The authors studied 100 families in which a fetus suspected of having the hemolytic disease was born, 70 women who had given birth to one or more stillborn fetuses and 60 mothers and their infants thought to have "physiologic jaundice." In the first group 97 of the mothers were Rh negative and 3 Rh positive. Serums of the Rh-negatiye mothers contained anti-Rh agglutinins in 93 out of 97 instances. Seventy-nine affected infants of Rh-negative mothers were tested, and all were Rh positive. Forty-five husbands of Rh-negative mothers tested were all Rh positive. The serums of all 3 Rh-positive mothers contained immune agglutinins incompatible with the fetal erythrocytes. In 1 instance the agglutinin was anti-Rh_2 and in the other 2 the agglutinin was anti-B. When several children were born to sensitized mothers, it was noted that the disease did not necessarily become more severe in successive siblings. In 5 families the birth of an affected infant was followed by the birth of 1 or more normal children. If any of the living children were Rh negative, the prognosis for future siblings appeared to be better.

Of the 70 women who had given birth to one or more stillborn fetuses, 17 were Rh negative and 53 Rh positive. In 11 cases the mother's serum contained immune anti-Rh agglutinins, and in all cases the fetus was stillborn during the last three months of pregnancy. It was concluded that in these cases the fetal death was due to hemolytic disease, while in the remainder this possibility could not be excluded.

Among the group of 60 infants affected with "physiologic jaundice," the mother's serum contained anti-Rh agglutinins in 3 cases out of 60, and in these cases the infant was Rh positive. In only 1 instance did the clinical examination suggest that the infant might have mild hemolytic disease. Compatibility tests between the mother's serum and the infant's erythrocytes with regard to the anti-A and anti-B agglutinins

94. Wiener, A. S.: Role of the Subtypes of Rh in Hemolytic Transfusion Reactions and in Erythroblastosis, Am. J. Clin. Path. **14**:52, 1944.

95. Boorman, K. E.; Dodd, B. E., and Mollison, P. L.: The Incidence of Haemolytic Disease of the Foetus (Erythroblastosis Foetalis) in Different Families: The Value of Serological Tests in Diagnosis and Prognosis, J. Obst. & Gynaec. Brit. Emp. **51**:1, 1944.

showed that the incidence of incompatibility in this group was not significantly greater than in a random sample of mothers and infants. Cases of mild hemolytic disease of the newborn may sometimes be mistaken for cases of "physiologic jaundice," however, unless serologic tests are carried out.

McCall, Race and Taylor [96] studied an infant with hemolytic disease of the newborn whose mother was Rh positive. The hemolysis was found to be due to a rare type of Rh antibody similar to the anti-Rh factor described by Levine and his collaborators.

Erythroblastosis fetalis in identical twins is reported by Demy.[97] A woman 40 years of age had been pregnant seven times. The first four pregnancies had resulted in living infants. There followed the birth of twins, 1 stillborn and the other dying of hemorrhage in four hours, and two years later a stillborn child thought to have had fetal hydrops was delivered. Erythroblastosis was suspected during the last gestation, and investigation showed the father's blood to be Rh positive and the mother's blood Rh negative in high titer. Labor resulted in the spontaneous delivery of identical twin girls. Severe hemolytic anemia with deep jaundice, petechial hemorrhages and a great enlargement of the liver and spleen developed in 1 infant. The other infant had mild anemia, light jaundice and a small percentage of normoblasts in the peripheral blood. Both were given transfusions of Rh-negative blood and recovered, although pigmented skin and an enlarged liver and spleen were still present in the severely afflicted twin fifty-five days after birth. Both infants were Rh positive. Being identical twins, they had identical genetic inheritance and were subject to the same titer of antibody from the mother. Demy explains the differing degree of severity of the anemia in the 2 infants as due to a greater functional defect with regard to passage of antibodies in the placenta serving the more severely affected twin.

Keel [98] summarizes the clinical findings in 12 cases of anemia neonatorum with icterus gravis. Four of the infants died and were studied pathologically. Extramedullary hemapoiesis was a constant finding. A group of 21 cases of hemolytic disease of the newborn were investigated by Langley and Stratton.[99] Previous observations in regard to the role of the Rh factor in this condition were confirmed by finding that in 19 of 21 cases the maternal blood contained an anti-Rh agglutinin. In 12 fatal cases necropsies were done, and in 11 of them extramedullary hemopoiesis was found. The breast milk of 10 of the mothers was examined, and anti-Rh agglutinins were found in the milk of 7.

Van Dorsser, Morrison and Philpott,[100] conducting a study of the significance of the Rh factor in obstetric practice, found that the percentage of abortions, miscarriages and fetal deaths among Rh-negative women was 21 per cent, as compared with 13 per cent among Rh-positive women. The percentage of Rh-negative and of Rh-positive women having no obstetric mishaps, however, were approximately equal. The Rh-negative women during their first two or three pregnancies at least were not particularly more apt to lose their children either before or during birth or in the neonatal period than were Rh-positive women. The incidence of toxemia was about the same in the two groups.

Adelman [101] reports on a pair of dissimilar twins, in 1 of whom severe anemia with jaundice, erythroblastosis, splenomegaly and hepatomegaly developed when the child was 48 hours old. By the use of repeated transfusions the blood values could be maintained, but the infant's clinical condition appeared to become steadily worse. Splenectomy was performed on the thirteenth day, and thereafter an uneventful recovery occurred.

Six infants with hemolytic anemia of the newborn are reported on by Lubinski, Benjamin and Strean.[102] All the mothers were Rh negative, one of them apparently having been sensitized earlier by transfusions of Rh-positive blood. The authors recommend in treatment the early use of washed Rh-negative red cells. Weiss [103] reviewed the accumulated knowledge concerning the clinical importance of the Rh antigen. He was able to study 15 families in which children with erythroblastic anemia had occurred and others in which the Rh antibody was the cause

96. McCall, A. J.; Race, R. R., and Taylor, G. L.: Rhesus Antibody in Rh-Positive Mother Causing Haemolytic Disease of Newborn, Lancet **1**:214, 1944.

97. Demy, N. G.: Erythroblastosis Fetalis in Identical Twins, Am. J. Obst. & Gynec. **47**:554, 1944.

98. Keel, M.: Anaemia Neonatorum and Icterus Gravis, Ann. pædiat. **160**:179, 1943.

99. Langley, F. A., and Stratton, F.: Hemolytic Disease in the Newborn, Lancet **1**:145, 1944.

100. van Dorsser, G. J. E.; Morrison, A. W., and Philpott, N. W.: Studies on Rh Factor and Its Significance in Obstetric Practice, Canad. M. A. J. **50**:219, 1944.

101. Adelman, M.: Hemolytic Disease of the Newborn (Erythroblastosis Foetalis) in One of Twins, Rhode Island M. J. **27**:580, 1944.

102. Lubinski, H.; Benjamin, B., and Strean, G. J.: Observations on the Rh-Factor in Its Relation to Hemolytic Anemia of the Newborn Infant, Am. J. Obst. & Gynec. **48**:464, 1944.

103. Weiss, C.: Rh Antigen: Its Clinical Importance, California & West. Med. **60**:59, 1944.

of transfusion reactions or was of interest in medicolegal connections.

Butler, Danforth and Scudder [104] report a serious post-transfusion reaction due to Rh antibody sensitivity. The patient was a 36 year old woman who had given birth to four living children and then had had four spontaneous miscarriages. She became pregnant again, and after five months' gestation fetal movements ceased. A diagnosis of fetal death was made. Two months later, active labor started and a macerated fetus was expelled. After delivery, uterine bleeding continued which could not be controlled by ordinary measures and the patient went into mild shock. A transfusion of Rh-positive blood compatible by the usual cross-match method was started and 100 cc. given before it was found that the patient was Rh negative. The blood transfusion was stopped immediately and plasma substituted. Forty minutes later the patient had a shaking chill. Bloody urine was passed. The hematocrit value dropped rapidly to nearly half of the original level, and the blood plasma showed hemolysis. The patient became jaundiced and generally edematous. The liver gradually increased in size until its lower edge lay 8 cm. below the right costal margin. The cephalin flocculation reaction was then 2 plus.

The patient was given sodium bicarbonate, transfusions of Rh-negative blood and plasma. Extreme oliguria persisted for seven days; nitrogen was retained until the nonprotein nitrogen concentration was 69.2 mg. per hundred cubic centimeters; the blood pressure became elevated, and a number of convulsions occurred. Recovery followed.

Investigation showed that anti-Rh agglutinins were present in high titer. The authors advise that blood transfusions never be given to obstetric patients until their Rh type is known.

Howell and Hobbs [105] describe a severe transfusion reaction which occurred in a 45 year old army officer who was given a blood transfusion after an operation for incision and drainage of a deep staphylococcus abscess of the thigh. He had had two blood transfusions about five years previously within a period of two months, and after the second of these he had had a severe chill and fever. The possibility of Rh incompatibility was considered when the third transfusion was given, but no typing serum was available. He was given 400 cc. of citrated blood over a period of ninety minutes without immediate reaction. Forty-five minutes later, however, he had a severe chill, which was followed by a rise in temperature to 102 F. He soon became jaundiced, and during the twenty-four hours following the blood transfusion he voided only 90 cc. of urine, containing large amounts of urobilinogen. There was nitrogen retention for several days, but a slow recovery ensued over a period of several weeks. When Rh typing serum was obtained, it was found that the recipient's cells were Rh negative and Rh positive agglutinins were demonstrated in his serum. The donor's cells were Rh positive. The authors emphasize the importance of determining the Rh factor in recipients of repeated blood transfusions, even though the interval may be of several years.

Elliptocytosis.—The great majority of persons who have the hereditary anomaly of oval or elliptic erythrocytes are healthy persons, long lived, not anemic and not unusually subject to disease. Several years ago Leitner investigated a large family in which elliptocytosis occurred as a benign deformity of the red blood cells. He has more recently [106] encountered another family in which the trait occurred in association with manifestations of hemolytic anemia, and he reviews a fairly large number of other reported cases in which this has occurred. In the family he studied 7 of the 8 members showed elliptocytosis, which was inherited as a dominant trait affecting both the males and the females of three generations. The erythrocyte deformity was present in fresh preparations, and there was no sickling or alteration in shape with exposure to carbon dioxide. In the 2 persons in whom the erythrocyte deformity was greatest, there was evidence of moderately severe hemolytic anemia with decreased resistance of the erythrocytes to hypotonic sodium chloride solutions, increased bilirubin in the blood and increased urinary urobilinogen. In 1 person the reticulocytes were increased to 38 per cent. There was no splenomegaly observed in any person.

Helz and Menten [107] report a case of elliptocytosis in a 2½ month old Negro child whose father had also had the abnormally shaped red blood cells. In the infant the percentage of oval cells gradually increased to 50 by the age of 4 months, after which no further increase was observed. Tests showed no signs of sickling.

104. Butler, B. C.; Danforth, D. N., and Scudder, J.: The Rh Factor in Intragroup Blood Transfusion Reactions, Surg., Gynec. & Obst. **78**:610, 1944.

105. Howell, R. P., and Hobbs, R. E.: A Hemolytic Transfusion Reaction Due to the Rh Factor, Mil. Surgeon **94**:269, 1944.

106. Leitner, St. J.: Weitere Untersunchungen über die familiäre Elliptozytose der Erythrozyten: Elliptozytose und hämolytische Anämie, Helvet. med. acta **10**: 585, 1943.

107. Helz, M. K., and Menten, M. L.: Elliptocytosis: Report of Two Cases, J. Lab. & Clin. Med. **29**:185, 1944.

Evans [108] studied a Nigerian native in whose blood 94 per cent of the erythrocytes were oval.

Sickle Cell Anemia.—Several authors have contributed studies during the last year showing the wide distribution of the sickling phenomenon among the dark-skinned races. Evans [109] performed tests for sickling on a group of 561 soldiers, natives of the Gambia, the Gold Coast, Nigeria and the Cameroons. Twenty per cent of the entire group had blood which showed sickling in vitro. Of the physically fit soldiers, 15.5 per cent showed sickling, in comparison with 25 per cent without sickling among those under treatment for various illnesses. A small group of villagers were examined, and the blood of 18.8 per cent showed sickling. A sex difference was noted in this group, since 22 per cent of the males showed sickling but only 13 per cent of the females. No cases of anemia associated with the trait were seen. English [110] reports the presence of the sickle cell trait in a young Bantu man, a native of North Rhodesia.

Mera [111] studied all the school children between the ages of 7 and 17 in Puerto Trejada, a Colombian town with an 80 per cent Negro population. Of the 489 Negro children examined, 46 (9.4 per cent) showed either the sickle cell trait (5.45 per cent) or sickle cell anemia (3.95 per cent). Typical symptoms of sickle cell anemia in these children included anemia, jaundice, chronic weakness, abdominal colic and ulcers of the leg.

The prevalence of sicklemia in the black Carib Indians, a Negro of relatively pure strain, was studied by McGavack and German.[112] The authors made wet, sealed films of blood from 300 persons living in a small village. Eight per cent of the persons who were studied showed the sickle cell trait. No sickle cell anemia or acute illnesses suggestive of the crises of sickle cell anemia were recognized in these persons.

Castellanos Fonseca [113] reviews much of the literature concerning the sickle cell disease and its general clinical features. Six cases of sickle cell anemia were reported in detail; in 2 the patients were Negroes and in 4 persons of mestizo origin. Investigation of 1 of the Negro families showed the sickle cell trait to be present in at least 10 males and females in three successive generations.

Triñcão and Rôlo [114] review the literature on sickle cell anemia and report on a white Portuguese family in which there were 5 cases in three generations. There was no admixture of Negro blood for at least five generations.

Urteaga Ballón [115] studied the value of splenic puncture as a diagnostic method in cases of latent sickle cell anemia with splenomegaly. In material aspirated from the spleen in such cases large numbers of deformed erythrocytes were present, and the procedure was recommended as an aid in diagnosis.

The behavior of erythrocytes from a patient with sickle cell anemia was studied by Murphy and Shapiro [116] under various experimental conditions. A method of making fresh blood preparations using an oiled slide was developed for the rapid demonstration of sickling. The effect on the cells of different serum and saline suspensions was studied, and sickling was found to occur more rapidly and more completely in unaltered whole fresh blood than in either serum or saline suspensions. Experiments confirming the fact that sickling depends on the state of the hemoglobin as regards oxygenation were described. When the hemoglobin was in the reduced form, the sickle shape of the erythrocyte was stable, but with the hemoglobin in oxygen combination the cell reverted to its normal shape. Sickling did not appear to be related to p_H or to variations in ionic balance. The potassium content of the erythrocytes, however, appeared to be significantly less than normal, and a preliminary test showed that sickle cells were nearly twice as permeable to the potassium ion as normal cells. The cells of different patients were found to show sickling at different thresholds of oxygen tension. The authors suggest that this phenomenon may be made the basis of a differential test between the sickle cell trait and the sickle cell disease. There was evidence to indicate that as the cells age the sickling threshold becomes lower, which is of possible relation to the genesis of the crises in sickle cell anemia.

108. Evans, W.: Elliptical Erythrocytes, J. Path. & Bact. **55**:378, 1943.

109. Evans, R. W.: The Sickling Phenomenon in the Blood of West African Natives, Tr. Roy. Soc. Trop. Med. & Hyg. **37**:281, 1944.

110. English, R. B.: Occurrence of Sickle Cell Trait in Blood of Bantu, South African M. J. **17**:389, 1943.

111. Mera, B.: Preliminares del estudio de la meniscocitemia en Colombia, S. A., Bol. Ofic. san. panam. **22**:680, 1943.

112. McGavack, T. H., and German, W. M.: Sicklemia in Black Carib Indian, Am. J. M. Sc. **208**:350, 1944.

113. Castellanos Fonseca, E.: Hemodistrofias por hematies falciformes, Rev. méd.-quir. de Oriente **4**:239, 1943.

114. Triñcão, C., and Rôlo, J.: Anemia de células falciformes; revisão da literature e apresentação de casos pessoais em brancos portugueses, Lisboa méd. **19**: 471, 1942.

115. Urteaga Ballón, O.: Importancia de la punción esplénica en el diagnóstico de la sickell cell anemia, Rev. med. exper., Lima **2**:177, 1943.

116. Murphy, R. C., Jr., and Shapiro, S.: Sickle Cell Disease: Observations on Behavior of Erythrocytes in Sickle Cell Disease, Arch. Int. Med. **74**:28 (July) 1944.

Winsor and Burch [117] observed that the sedimentation rate in sickle cell anemia was increased by exposing the blood to oxygen and reduced by exposing the blood to carbon dioxide. They describe two methods of making the tests and present data showing that the induced changes in sedimentation rate were constant enough to be of value in diagnosis. The test could be completed in less than thirty minutes in many cases.

The same authors [118] studied a group of 15 patients with sickle cell anemia and 4 patients with the sickle cell trait. Anthropometric measurements, photographs and roentgenograms were compared with the clinical histories and other data. A characteristic habitus, not a primary genetic abnormality but an alteration secondary to the disease, was found in those patients with severe active sickle cell anemia of long duration. Patients with sicklemia only showed no significant deviation from control subjects.

Sickle cell anemia is a disease that is frequently of surgical interest because the crises may simulate acute surgical diseases of the abdomen. Bauer and Fisher [119] report several fatal cases of sickle cell disease, most of them not correctly diagnosed during the life of the patient, and urge that routine tests for sickling be made on all Negro patients requiring admission to a hospital. Canby, Carpenter and Ellmore [120] report the case of a 19 year old youth of pure Sicilian stock who became acutely ill with abdominal pain in the left side, fever and leukocytosis. He had been pale most of his life, and some yellowness of the scleras had often been noted. A laparotomy was thought advisable, and an enlarged congested spleen covered with adhesions and fibrin was found and removed. The convalescence was uneventful. His erythrocytes showed complete sickling in twelve hours. There was no sickling in the blood of either parent, four sisters or two brothers after twenty-four hours.

Reinhard and his collaborators [121] used oxygen therapy in the treatment of sickle cell anemia. Four patients breathed 70 to 100 per cent oxygen administered through a Boothby-Lovelace-Bulbulian mask for six periods of eight to twenty days each. They observed that oxygen therapy resulted in a decrease in the degree of intravascular sickling of the red blood cells but that there was no consistent detectable change in the rate of hemolysis. Erythrocytogenesis was depressed by the oxygen therapy. On the fourth or fifth day of oxygen therapy the percentage of reticulocytes began to decline, and this decline was followed in two or three days by a fall in the number of erythrocytes, which was occasionally as much as 1,000,000 cells per cubic millimeter. With discontinuation of the oxygen inhalation, a striking reticulocytosis developed and the level of erythrocytes returned to the preoxygen level. No definite conclusions as to the value of oxygen therapy for the crises of sickle cell anemia were drawn.

MEDITERRANEAN ANEMIA

Several new studies of Mediterranean anemia have appeared during the past year, showing that anemias of this type are less uncommon and of wider geographic and racial distribution than is generally considered. Among the natives of Cyprus, Fawdry [122] found that erythroblastic, or Cooley's, anemia was a common disease among the children and young adults. He studied 20 patients ranging in age from 6 months to 20 years, all of them, with the exception of 1 Turkish boy, being of Greek ancestry. The general clinical aspects and the conditions observed in the blood and bone marrow in his cases were in agreement with previous studies by other investigators. The use of iron and liver preparations as well as of antimalarial drugs was of no therapeutic value.

Saracoglu [123] reports the typical features of Cooley's anemia in a 9 year old Turkish boy. A brother of the patient had had a greatly swollen abdomen and died of anemia at the age of 5 years. An uncle of the patient was thought to be anemic also. The author stated that his patient was the fourth to be reported on among the Turkish people. Dhayagude [124] reports on

117. Winsor, T., and Burch, G. E.: Diagnostic Physicochemical Blood Tests in Sickle Cell Anemia, Am. J. M. Sc. **207**:152, 1944.

118. Winsor, T., and Burch, G. E.: The Habitus of Patients with Sickle Cell Anemia, Human Biol. **16**: 99, 1944.

119. Bauer, J., and Fisher, L. J.: Sickle Cell Disease, with Special Regard to Its Nonanemic Variety, Arch. Surg. **47**:553 (Dec.) 1943.

120. Canby, C. B.; Carpenter, G., and Ellmore, L. F.: Drepanocytosis (Sicklemia) and an Apparently Acute Surgical Condition of the Abdomen: Report of Their Occurrence in a White Youth with Laparotomy, Arch. Surg. **48**:123 (Feb.) 1944.

121. Reinhard, E. H.; Moore, C. V.; Dubach, R., and Wade, L. J.: Depressant Effect of High Concentrations of Inspired Oxygen on Erythrocytogenesis: Observations on Patients with Sickle Cell Anemia with a Description of the Observed Toxic Manifestations of Oxygen, J. Clin. Investigation **23**:682, 1944.

122. Fawdry, A. L.: Erythroblastic Anaemia of Childhood (Cooley's Anaemia) in Cyprus, Lancet **1**:171, 1944.

123. Saracoglu, K.: Cooleysche Anämie in der Türkei, Wien. med. Wchnschr. **93**:217, 1943.

124. Dhayagude, R. G.: Erythroblastic Anemia of Cooley (Familial Erythroblastic Anemia) in an Indian Boy, Am. J. Dis. Child. **67**:290 (April) 1944.

a 7 year old Brahmin boy, who also showed the blood and skeletal abnormalities typical of severe Mediterranean anemia. Erythrocyte fragility tests and determinations of blood levels of bilirubin on 2 half brothers and a half sister showed them to be normal. The author stated that his patient was the fourth to be observed among Asiatic persons.

A comprehensive study of the hematologic features and the genetic transmission of Mediterranean anemia, or thalassemia, is reported by Valentine and Neel.[125] Hematologic examinations were made on the parents, the siblings and some immediate collaterals of 3 persons who had severe Mediterranean, or Cooley's, anemia and of 1 person with a similar milder condition. The four families included a total of thirty-four persons. Twenty-four of these persons had the mild, benign form of anemia described in recent years by many authors as occurring among families of Mediterranean origin. The mild anemia was qualitatively similar but quantitatively less severe than the full-blown thalassemia and was characterized by increased resistance of the erythrocytes to hemolysis in hypotonic sodium chloride solutions, target and oval red blood cells, microcytosis and hypochromia. All patients with the Mediterranean disease, in the authors' opinion, can be roughly divided into two groups: those with mild, usually asymptomatic anemia and those with severe anemia usually terminating fatally in childhood. It is proposed that these two forms of the disease be designated "thalassemia minor" and "thalassemia major," respectively.

In two families in which there had been children with the severe anemia, or thalassemia major, both parents were available for hematologic examination, and in each family both parents were shown to have significant hematologic abnormalities. In reviewing the literature, the authors found that in every family in which the parents of children with the severe Mediterranean anemia have had complete blood examinations, with 1 possible exception, definite hematologic abnormalities have been demonstrable in both parents. Several possible genetic mechanisms in the heredity of thalassemia have been suggested in the past. The authors believe that it is most likely that thalassemia minor is due to heterozygosity for a factor which when homozygous results in thalassemia major. The problem of the detection of the carriers of thalassemia and the differential diagnosis of the mild anemia are discussed.

Voorhies and Sloan[126] report the case of a 20 year old youth of Sicilian parentage, who was found, while serving in the United States Army, to have a mild form of Mediterranean anemia. While he was being treated for a poison ivy dermatitis, it was discovered that he was slightly icteric and that his spleen was enlarged, with the tip extending to the level of the umbilicus. Except for a tendency to have frequent epistaxes, the inability to run because of pain in the upper left part of the abdomen and shortness of breath on exertion, his general health had been good. There was a moderate degree of anemia present, and the red blood cells varied excessively in size and shape. Ten to 15 per cent of them were target cells. Roentgenograms showed thickening of the calvarium and in other bones osteoporosis with widened trabeculae. Corcoran[127] observed the skeletal abnormalities characteristic of erythroblastic anemia in a roentgenogram of an 8 year old Italian boy who had fractured his femur. Further roentgen study showed the osseous changes to be generalized. The fractured bone healed normally. Novak[128] discovered a number of dental abnormalities as well as prominent trabeculae in roentgenograms of the maxillary and mandibular bones of a child with erythroblastic, or Cooley's, anemia.

TOXIC EFFECTS OF INDUSTRIAL POISONS

A general article discussing benzene as a toxic hazard in industry is written by Browning.[129] She reiterates that the extensive use of benzene in industry is largely due to the fact that it is an excellent solvent for many substances, such as rubber, nitrocellulose, fat, oils and resins. Unfortunately, despite efforts to prevent toxic effects, they are not infrequently observed, especially in the nervous system and the bone marrow. Hence benzene poisoning is a potential risk to health wherever the substance is used in significant amounts. According to the author, there are two separate fields of application of benzene in industry, each of which carries its own distinct, and to some extent different, risks to health. The first is distillation of coal and

125. Valentine, W. N., and Neel, J. V.: Hematologic and Genetic Study of the Transmission of Thalassemia (Cooley's Anemia; Mediterranean Anemia), Arch. Int. Med. **74**:185 (Sept.) 1944.

126. Voorhies, N. W., and Sloan, F. R.: Mediterranean Anemia, J. A. M. A. **125**:352 (June 3) 1944.

127. Corcoran, W. J.: Erythroblastic Anemia, with Report of a Case in a Boy 8 Years Old, Radiology **43**: 373, 1944.

128. Novak, A. J.: The Oral Manifestations of Erythroblastic (Cooley's) Anemia: Case Report, Am. J. Orthodontics **30**:539, 1944.

129. Browning, E.: Benzene as a Toxic Hazard to Industry, Brit. J. Phys. Med. **7**:122, 1944.

coal tar, in which the risk is chiefly that of acute poisoning or "gas." This most frequently arises from accidental leakages or breakages or from the failure of the worker to take full precautions when entering vats or tanks for the purpose of cleaning them. The second is the use of benzene as a solvent or a diluent, as in the rubber and lacquer industries, airplane "doping" and manufacture of artificial leather, in which the risk is mainly that of chronic poisoning, with its attendant symptoms and hematologic changes. Benzene poisoning, therefore, results either from absorption from the skin or from inhalation, and it is imperative that contact by either of these routes should be reduced to a safe level. A diagnosis of chronic benzene poisoning cannot be made on the evidence of symptoms alone, for hematologic changes do not by any means always run parallel with the severity of subjective symptoms nor are either the hematologic changes or the damage to the bone marrow invariably characteristic. Although aplastic anemia is the most frequent outcome of severe benzene poisoning, there have been cases in which the final condition is that of leukemia, in which the bone marrow has been found after death in a state of hyperplasia or of combined hyperplasia and aplasia. According to the author, however, it is clear that the earliest signs of benzene poisoning is a reduction of those elements of the blood stream which are believed to provide the body with its powers of resistance to infection, namely, the polymorphonuclear leukocytes. Even though the white blood cell count is 5,000 to 6,000 per cubic millimeter, the differential count of the stained smear in affected patients will show change in the proportion of polymorphonuclear leukocytes, so that the total number of these cells is below 3,000 per cubic millimeter. If the workers are removed from the exposure to benzene at this stage, there is every reason to believe that the depressed bone marrow will recover and that no permanent ill effects will remain. When the damage has progressed to the stage of true aplastic anemia, blood transfusions and injections of pentnucleotide are sometimes successful, but if the degree of aplasia is extreme, no treatment seems to be efficacious. The important observation is emphasized that all persons who come into contact with benzene are not equally susceptible to its toxic effects. Among a large number of workers who are equally exposed for the same length of time and to the same concentration, only a few may have any symptoms of ill health or show any characteristic hematologic change. On the other hand, extremely severe poisoning may arise in any susceptible person with only moderately severe exposure.

A report of a case of fatal poisoning with benzene is given by Ackerly and Hawlick.[130] The patient was a man 35 years old, who was exposed to benzene as a result of rinsing glassware with it six to eight times a day. This required ten to fifteen minutes for each operation. When the patient was admitted to the hospital, his hemoglobin content was 34 per cent, red blood cell count 1,550,000 per cubic millimeter, with 3.7 per cent reticulocytes, and white blood cell count 3,450 per cubic millimeter, with 10 per cent stab forms, 40 per cent segmented forms and 50 per cent lymphocytes. The platelet count was 171,000 per cubic millimeter, bleeding time three minutes ten seconds and coagulation time three minutes forty-five seconds. Despite all treatment, the patient died. Observations at necropsy showed, among other things, reduction in the total number and irregularity in the distribution of the myeloid elements. The erythroblastic elements were represented by small clumps of dark-staining cells, chiefly normoblasts. Megakaryocytes were numerous and irregularly spaced, and there was pronounced variation in their size, shape and nuclear configuration. All the myeloid elements were present, but mature granulocytes were less numerous than normal.

A report on a patient in whom thrombocytopenic purpura due to benzene poisoning developed is presented by Vaughan,[131] and the literature bearing on this subject is discussed. He makes the point that depressed production of platelets is a commonly associated phenomenon in benzene poisoning but that cases of thrombocytopenic purpura unassociated with striking changes in other marrow elements are rarely found. The patient observed by Vaughan was a woman 33 years of age, whose chief complaint was menorrhagia of nine months' duration and purpura of sixteen months' duration. She was first seen by a surgeon, who performed a dilation and curettage and finally implanted a radium bomb for eighteen hours. A short time later the patient was admitted to the medical service of the hospital and showed the classic hematologic features of thrombocytopenic purpura. The platelet count was 20,000 per cubic millimeter, bleeding time eighteen minutes, clotting time eleven minutes and clot retraction 47 per cent of

130. Ackerly, R. H., and Hawlick, G. F.: Benzene (Benzol) Poisoning: Report of a Fatal Case with Autopsy Findings, Rocky Mountain M. J. **41**:402, 1944.

131. Vaughan, W. T., Jr.: Thrombocytopenic Purpura Due to Benzol Poisoning, J. Indust. Hyg. & Toxicol. **26**:274, 1944.

normal. The tourniquet test resulted in 159 purpuric spots to the square inch, with 10 spots in the control. The prothrombin time was fifty-five seconds (the control was forty-five seconds) and the ascorbic acid level was normal. Aspiration of sternal marrow revealed an essentially normal marrow picture with no increase in megakaryocytes. The patient reported that for sixteen years, between the ages of 14 and 31, she had worked in various shoe factories as a "cementer." At this time she handled all kinds of cement, which dried on her hands in the course of her work. Just prior to the first episode of purpura, however, the patient had terminated a four month period of cementing in a factory reported not to be using benzene as a solvent. Vaughan estimates that the interval between exposure and the development of clinical symptoms in his patient was approximately six to twelve months. Whereas undoubtedly benzene must be considered as an important etiologic factor in the production of purpura in this patient, it is not proved beyond the question of a doubt that it was the responsible cause. The case emphasizes, however, the care with which the history must be taken in all cases of diseases of the blood which might be attributable to the effects of various industrial poisons or toxins. A further follow-up study of this patient would be of extreme interest, to learn whether some other disease condition, such as a subleukemic leukemia or true idiopathic thrombocytopenic purpura, might develop.

Cone [132] reviews the literature concerning the effect of trinitrotoluene (TNT) on the blood and studied a group of workers exposed to this chemical in a munitions industry. Weekly blood counts were performed on 17 workers in intimate contact with trinitrotoluene for eight weeks and then monthly for an additional four months. During the period of study the trinitrotoluene concentration of the air was determined and found to be consistently below the proposed threshold level of 2.5 mg. per cubic meter of air. In all there was relatively little change in the erythrocyte count during the investigation, and there were no changes in the size and shape of the red blood cells. The hemoglobin level was not affected. Most of the persons studied showed a transient initial rise of leukocytes the count usually varying from 11,000 to 12,000 per cubic millimeter, and there was a moderate eosinophila, but in some instances the proportion of eosinophils was as high as 15 per cent. The hematologic changes were not accompanied with symptoms. No other alterations were noted in the formed elements of the blood.

Three fatal cases of trinitrotoluene poisoning are reported by McNally.[133] He reviews the literature and points out that trinitrotoluene may gain entrance into the blood stream by absorption through the skin, lungs or gastrointestinal tract. Unless masks are worn, fine dust and fumes will reach the mucous membranes of the nose and mouth and be swallowed with the saliva and mucous secretions. There seems to be general agreement that the skin is the most important portal of entry. He states that the outstanding features of trinitrotoluene poisoning are the changes found in the blood, jaundice, degenerative hepatitis (the liver being reduced to one half of its normal weight) and the renal damage. The hematologic changes are described as varying considerably, depending on the intensity of the exposure and the length of time the person has been employed on the particular job. Sometimes there is extensive anemia, with a hemoglobin content reported as low as 28 per cent and a red blood cell count of 1,225,000 per cubic millimeter, with a variation of the white blood cells from 2,800 to 14,650. According to McNally, two theories have been advanced as to the origin of the anemia. Some investigators believe the hepatic injury to be the primary source, while others assume that the destruction of the red blood cells is the cause of the icterus and that it is therefore of hemolytic origin.

The uses of tetrachloroethane in industries associated with war activities are described by Coyer,[134] and 7 cases of poisoning with this chemical are described. It is a colorless, mobile liquid with an odor resembling that of chloroform and carbon tetrachloride, is nonflammable and has been used as a solvent for various substances, such as cellulose acetate and nitrate, resins, waxes, pitch and various other materials. It is the most toxic of the chlorinated hydrocarbons and produces not only a narcotic effect but profound metabolic injuries, especially atrophy of the liver. Reference to the original article should be made for details of the toxic effect. The changes in the blood are not striking. There usually is a moderate reduction in the red blood cell count, to the vicinity of about 3,500,000 red blood cells per cubic millimeter, a moderate increase of leukocytes, the count varying from

132. Cone, T. E., Jr.: Review of Effect of Trinitrotoluene (TNT) on Formed Elements of Blood, J. Indust. Hyg. & Toxicol. **26**:260, 1944.

133. McNally, W. D.: Three Deaths from TNT (Trinitrotoluene) Poisoning, Indust. Med. **13**:491, 1944.

134. Coyer, H. A.: Seven Cases of Tetrachloroethane Poisoning and Review of Several Cases Treated, Hahneman. Monthly **79**:230, 1944.

11,000 to 13,000 per cubic millimeter and no other changes of importance in the blood.

It has been found by Van Loon and Clark [135] that the administration of acetanilid up to 36 mg. per kilogram per day over long intervals caused little or no abnormal hematologic changes in dogs. Larger doses of the drug, however, produced a hemolytic anemia but with no evidence of depression of the bone marrow. Recovery was rapid and apparently complete on cessation of administration of the drug. The same effect was produced by the administration of acetophenetidin, but quantitatively the drug is less active than acetanilid. A temporary methemoglobinemia is produced by both drugs when certain threshold doses are exceeded. In the range of the dose employed, however, no accumulation of methemoglobin was observed in repeated daily administrations.

APLASTIC ANEMIA

A general discussion of aplastic anemia is given by Pearce,[136] in which he makes the following main points: First, in all cases of anemia in which the bone marrow shows signs of regeneration there should be a thorough search made for the offending agent and, if possible, it should be eradicated. Second, not all the diagnostic criteria are present in every case of early aplastic anemia, since depression of granulocyte or thrombocyte formation may not develop until some time after depression of erythrocyte formation. He also properly emphasizes that even signs of regeneration of erythrocytes may be apparent at times as evidenced by a patchy condition of the marrow. Third, he insists that treatment should be persistent. It is his opinion that as long as there is life there is hope that a plastic or even aplastic marrow may ultimately become regenerative. Fourth, he emphasizes that repeated blood transfusions are still the main source of treatment but that marrow transfusions should be given further trial. We have observed spontaneous remissions in the course of idiopathic aplastic anemia, and we therefore agree that treatment should be persistent. We have also seen, as a result of one hundred and thirty-eight blood transfusions administered to 1 patient over a course of eight years, the development of hemochromatosis, presumably resulting from the injection of the iron contained in the hemoglobin of the transfusions. This is a matter which should be kept in mind.

A patient with aplastic anemia is discussed by Halliday [137] in whom the characteristic blood picture was present. This patient had been treated for sore throat with intravenous injections of 2.1 Gm. of a French preparation of neoarsphenamine. Subsequently, owing to a recurrence of the inflammation, the patient received several courses of sulfapyridine and sulfathiazole. The red blood cell count was found, when determined for the first time after both forms of medication had been given, to be 3,100,000 per cubic millimeter; the hemoglobin content was 8.2 Gm. per hundred cubic centimeters, and the leukocytes numbered 2,700 per cubic millimeter, 5 per cent being neutrophils. Blood platelets were "scanty." The bleeding time was twenty minutes. Hemolytic anemia resulting from the untoward effects of the sulfonamide drugs is also discussed by Halliday. In general, he believes that the toxic reactions due to the sulfonamide compounds result from (*a*) ill considered administration, (*b*) careless management of the patient, (*c*) prolonged course of treatment (over ten days) and overdosage and (*d*) allergic idiosyncrasy and sensitization. In treating agranulocytosis, in addition to immediate withdrawal of the drug, he recommends repeated blood transfusions and injections of sodium pentnucleotide. In his opinion, the value of liver extract and preparations of bone marrow appears doubtful. In treating patients with thrombocytopenic purpura due to sulfonamide compounds, the suspension of the use of the drug and repeated transfusions are recommended. In treatment of hemolytic anemia, he favors repeated blood transfusions and use of alkalis and later oral administration of iron.

Fatal bone marrow intoxication following exposure to trinitrotoluene is described by Hart and his associates [138] and by Eddy.[139] The former report 4 cases and the latter reports 3, but apparently 1 case is included in both series. All the patients were employed in munition plants. The duration of exposure to trinitrotoluene varied from thirty days to eleven months. One of the patients, a 26 year old man, reported on by Eddy, was employed in the shipping room of a bomb line, where there was no demonstrable trinitrotoluene in the air. All the others in both series were exposed to concentrations of the chemical

135. Van Loon, E. J., and Clark, B. B.: Observations on Hematologic Actions of Acetanilid and Acetophenetidin in Dog, J. Lab. & Clin. Med. **29**:942, 1944.

136. Pearce, R. S.: Hypoplastic or Aplastic Anaemia, Memphis M. J. **19**:51, 1944.

137. Halliday, J. A.: Aplastic Anemia, M.J. Australia **1**:352, 1944.

138. Hart, W. L.; Ley, E. B.; Scroggie, V. D.; Johnson, E. A., and Eddy, J. H. Jr.: Report of Four Cases of Aplastic Anemia Occurring Among Munitions Workers, Indust. Med. **13**:896, 1944.

139. Eddy, J. H., Jr.: Aplastic Anemia Following Trinitrotoluene Exposures: A Report of Three Cases, J. A. M. A. **125**:1169 (Aug. 26) 1944.

ranging from 1 to 10.7 mg. per cubic meter of air. The clinical course was typical of myeloid aplasia, with anemia, hemorrhagic manifestations and intercurrent infection. The duration of symptoms varied in different patients from about two weeks to seven months. Therapy was of no avail, except for temporary benefit from blood transfusions, and death usually occurred as the result of hemorrhage or infection, although in 1 case a contributing factor appeared to be reaction to blood transfusions with hemolysis and hepatic damage. In all instances in which examinations of marrow were performed, a pronounced degree of hypoplasia was found affecting both the erythropoietic and the leukopoietic elements. Although Hart and his associates state that "the most dependable laboratory finding indicative of toxic reaction . . . is the reduction in the hemoglobin and red blood count," it is our opinion that the leukocyte count and neutrophil percentage constitute an earlier and more sensitive index of marrow intoxication.

Aplastic anemia complicating therapy with oxophenarsine hydrochloride for syphilis is reported by Freeman.[140] His case was that of a 37 year old soldier who had received treatment with neoarsphenamine irregularly over a period of two years with several febrile reactions but no other toxic manifestations. After an interval of approximately three months following the last injection of neoarsphenamine, treatment with oxophenarsine hydrochloride was begun. After the tenth injection of the latter drug purpura appeared, but, in spite of this, two more injections were given. The patient died nineteen days after he had first noted the purpura. Findings at necropsy included severe ulceronecrotic lesions throughout the mouth and upper respiratory tract, multiple hemorrhages in skin, mucous membranes and serous membranes and bilateral lobular pneumonia. There was aplasia of all cellular elements of the bone marrow. Two other previously reported cases of aplastic anemia following use of oxophenarsine hydrochloride are reviewed.

The diagnosis of osteosclerotic anemia, or the anemia associated with Albers-Schönberg disease, is discussed by Binder and Riedl [141] in reporting 2 cases. The diagnosis was suspected first in both instances when great difficulty was encountered in doing sternal punctures. The first patient was a 29 year old mason, who complained of weakness, shortness of breath and bleeding from his nose and gums for two months. The liver and spleen were found to be slightly enlarged. The hemoglobin content was 55 per cent, red blood cells 2,900,000, white blood cells 7,000, hematocrit reading 24 and average cell volume 82 cubic microns. The differential count included a few myelocytes and about 1 nucleated red blood cell for every 100 white blood cells. The reticulocytes were 55 per cent. There was severe thrombopenia. A sternal puncture was impossible, since the needle broke in the attempt to penetrate the hard bone. Roentgenograms were then obtained, which showed generalized increase in skeletal density and bony sclerosis. The patient finally died of a generalized tendency to bleeding. Necropsy revealed generalized overgrowth of bony and fibrous tissue, with obliteration of the marrow cavity. There was no definite evidence of extramedullary blood formation. The second patient was a 42 year old man who had been weak and anemic for six months following a febrile illness. Blood transfusions and injections of liver extract had been of little benefit. The erythrocyte count was 1,800,000, hemoglobin content 37 per cent, hematocrit reading 17 and average cell volume 94 cubic microns. Ten per cent of the white blood cells were myelocytes. The reticulocyte percentage was 6. Sternal puncture was attempted but could not be done because of the dense bone. Roentgenograms of the bones showed greatly increased density, thickening and sclerosis. The patient improved after a series of blood transfusions.

TOXIC EFFECT OF THE SULFONAMIDE DRUGS INCLUDING AGRANULOCYTOSIS

An informative and reliable article dealing with the toxicity of sulfonamide drugs is presented by Lyons and Van Harn.[142] The authors cite statistics to show that toxic manifestations, such as fever, rash, hemolytic anemia, leukopenia, agranulocytosis, hematuria, oliguria and hepatitis, are present in about 12 per cent of the patients receiving sulfanilamide, in 15.9 per cent receiving sulfapyridine, in 18.6 per cent receiving sulfathiazole and in 6.5 per cent receiving sulfadiazine. They state that experience with sulfamerazine and sulfamethazine is as yet insufficient to permit cleancut decisions concerning the incidence of toxicity of these drugs. There is evidence to indicate, however, that sulfamerazine has about the same toxicity as sulfadiazine. Their state-

140. Freeman, H. E.: Aplastic Anemia with Acute Agranulocytosis, Thrombopenic Purpura and Complicating Mapharsen Therapy, Arch. Dermat. & Syph. **50**: 320 (Nov.) 1944.

141. Binder, L., and Riedl, O.: Beiträge zur Diagnostik der osteosklerotischen Anämie, München. med. Wchnschr. **89**:519, 1942.

142. Lyons, R. H., and Van Harn, R. S.: Problems of Toxicity in Sulfonamide Therapy, J. Oral Surg. **2**: 118, 1944.

ment in regard to the hematologic disorders which result from sulfonamide therapy may be summarized briefly as follows: They emphasize that such disorders as agranulocytosis, acute hemolytic anemia and thrombopenic purpura are uncommon but exceedingly serious complications of this form of treatment. They are encountered less often with sulfathiazole therapy than with sulfapyridine or sulfanilamide therapy, a point which we believe requires confirmation.

It is not clear whether these untoward results are due to hypersensitivity or whether in some cases they may be a result of direct toxic effect of the sulfonamide drugs. According to Lyons and Van Harn, hemolytic anemia is most likely to occur from the third to the fifth day of treatment, and it appears to be an expression of an individual idiosyncrasy to the drug. A more slowly developing anemia associated with hemolysis of the erythrocytes is not infrequently seen with sulfanilamide therapy and especially with sulfapyridine therapy, but it rarely occurs with sulfathiazole or sulfadiazine therapy. It is usually not an important complication of treatment, since it can be easily controlled by transfusions.

It has been noted by the authors that thrombopenic purpura has followed the use of sulfathiazole and sulfadiazine as well as the use of the earlier sulfonamide compounds. They state that too few cases have been studied to be certain whether this is the result of a direct toxic effect on the bone marrow or of an individual idiosyncrasy to the drug. The authors point out that, in contrast to acute hemolytic anemia and thrombopenia, it is uncommon for leukopenia or agranulocytosis to occur early in the course of sulfonamide therapy. These complications most frequently develop between the fifteenth and twenty-fifth days of treatment. In some cases readministration of the drug causes a recurrence of the agranulocytosis, but not in all instances. They emphasize that there is a second type of leukopenia in agranulocytosis, perhaps related to the lowering of the intestinal synthesis of essential growth factors through the administration of succinylsulfathiazole or sulfaguanidine. In animals this type of agranulocytosis is benefited by the administration of a liver extract.

The toxic effects of sulfonamide drugs are discussed by van Dyke [143] in a symposium dealing with these drugs. He emphasizes among other untoward effects the frequency of leukopenia and agranulocytosis. He states that "probably there have been hundreds of fatalities." He believes that granulocytopenia or agranulocytosis is usually observed in patients who receive large total doses of sulfonamide drugs over periods of eighteen to twenty-five days. It is his opinion that the complication rarely appears earlier than twelve days after the beginning of treatment, which has also been borne out in our experience. He cites the case of 1 patient reported on in the literature, who received 54 Gm. of sulfanilamide during nineteen days and of another who received 44 Gm. of the drug in twenty-five days before this complication appeared. He recognizes that any one of the sulfonamide drugs may provoke granulocytopenia and agranulocytosis but that the appearance of the syndrome has been reported more frequently after the administration of sulfanilamide, perhaps because the total number of patients treated with this drug is greater than the number of patients treated with any other sulfonamide compound.

Van Dyke also discusses the occurrence of hemolytic anemia due to treatment with sulfonamide drugs. He believes that a drug like sulfanilamide probably causes this change more frequently in children than in adults. Hemolytic anemia may result from the use of sulfanilamide, sulfapyridine, sulfathiazole, sulfadiazine or diaminodiphenylsulfone. Rarely, in his opinion, does aplastic anemia develop from the use of these therapeutic agents. He believes that perhaps as satisfactory a hypothesis as any relating to the cause of hemolytic anemia is the one that it results from increased fragility of the erythrocytes, attributable to the oxidation products of the sulfonamide drugs. It is also mentioned by Van Dyke that thrombopenia with purpura is a rare complication of sulfonamide therapy, of which about half the reported cases have terminated fatally.

The toxic reactions in 1,936 patients treated with sulfathiazole, sulfadiazine, sulfapyrazine, sulfaguanidine and succinylsulfathiazole are reported by Vilter and Blankenhorn.[144] Toxic reactions were found to occur in 116 patients, an incidence of 6 per cent. These untoward manifestations were usually of such severity as to compel the arrest of treatment. Occasionally therapy could be resumed by changing to another drug, and this was most frequently done by substituting sulfadiazine for sulfathiazole. Death was ascribed solely to the toxic effects of these drugs in 4 instances (0.2 per cent), and in 5 other patients it was certainly hastened by drug intoxication. All fatal toxic reactions were mainly renal and resulted in uremia. Altera-

143. Van Dyke, H. B.: The Toxic Effects of the Sulfonamides, Ann. New York Acad. Sc. **44**:477, 1943.

144. Vilter, C. F., and Blankenhorn, M. A.: The Toxic Reactions of the Newer Sulfonamides, J. A. M. A. **126**:691 (Nov. 11) 1944.

tions in the blood and bone marrow were noted 11 times with the use of sulfathiazole, once with sulfapyrazine and not at all with sulfadiazine. Of the patients who received sulfathiazole, hemolytic anemia developed in 4, leukopenia in 5 and agranulocytosis in 2. All patients recovered. In 1 patient who received sulfapyrazine leukopenia developed. In a discussion of this article, Dr. S. L. Bernstein, of Cleveland, warns against prescribing sulfonamide drugs to ambulatory patients. He remarked that he had given a man aged 60 a small dose of sulfanilamide for septic sore throat. The patient received a total of 20 grains (1.3 Gm.) with alkali for three days, and agranulocytosis developed, from which he recovered. This seems to be a small amount of sulfanilamide to be responsible for the development of agranulocytosis, especially if the patient had not previously received the drug.

In reviewing the type and frequency of reactions in 186 cases of sulfonamide intoxication, it was noted by Frist [145] that leukopenia occurred in 2 of 33 patients given sulfanilamide, 2 of 16 patients given sulfapyridine and 2 of 71 patients given sulfathiazole. A "secondary anemia" occurred in 5 patients of the 33 receiving sulfanilamide, 1 of the 16 receiving sulfapyridine, 2 of the 60 receiving sulfadiazine and in 4 of the 71 receiving sulfathiazole. In a total of 186 cases, therefore, there were 12 cases of "secondary anemia" and 6 cases of leukopenia.

Kracke,[146] in discussing hematologic problems before the Section on General Practice of the Southern Medical Association in November 1943, stated that an "occasional patient" may show a depression of the bone marrow from the action of sulfonamide drugs to the extent that the white blood cell count is extremely diminished and that the condition of agranulocytosis may subsequently develop. He also stated that he had seen 6 patients in whom there had been a pronounced thrombopenia following the use of these drugs. He regarded this as a serious clinical problem, since 4 of his patients died from resulting uncontrollable hemorrhages.

A study of the toxic reactions which occurred in 1,357 hospital patients treated with sulfadiazine orally or sodium sulfadiazine intravenously is reported by Plummer and Wheeler.[147] Of this entire group, a definite drop in the number of white blood cells below 3,000 per cubic millimeter or in the percentage of granulocytes below 35 was observed in only 6 patients, but in none of these did the total white blood cell count fall below 2,000 per cubic millimeter or the percentage of granulocytes below 10. There were no changes in the red blood cells observed. In 1 of the surgical patients, however, a fatal thrombopenia occurred after six days of treatment with sulfadiazine in a dose of 6 Gm. daily. This was the only fatal toxic reaction in the group.

Three fatal cases of streptococcic infection treated with sulfadiazine are reported by Geever,[148] with complete autopsy observations. There were no changes in the peripheral blood or bone marrow observed in the patients. A good point is emphasized by Geever, however, with which we are in complete accord; namely, that if a decision to continue the use of the drug in treatment of any patient in whom a cutaneous eruption develops during sulfadiazine treatment is made, then toxic effects should be expected and weighed against any possible beneficial influence on the original infection. In other words, the presence of a cutaneous eruption is a definite warning that subsequent important complications, of which agranulocytosis is the most serious, may be observed if the medication is continued.

Three fatalities following the use of sulfonamide drugs are reported by Gessler,[149] and a review is given of 30 additional cases, with necropsies, which were reported in the literature. In 5 of the 30 cases, agranulocytosis was a complicating factor, twice being due to the use of sulfathiazole, twice to sulfapyridine and once to sulfanilamide. In no case reported in this group did the complication follow the use of sulfadiazine. In most instances the dose was 6 Gm. per twenty-four hours; the average daily dose never exceeded 10 Gm. From his study the author concludes that uremia and agranulocytosis are the most frequent lethal complications. Sulfathiazole, according to him, has more chance of causing urinary complications than have other drugs. He states that the microscopic picture of the bone marrow in cases of agranulocytosis due to sulfonamide intoxication is usually that of arrest of cell maturation. He quotes, however, from the article by Lederer and Rosen-

145. Frist, T. F.: Reactions to Sulfonamide Compounds: Review of One Hundred and Eighty-Six Cases, War Med. **5**:150 (March) 1944.

146. Kracke, R. R.: Hematologic Problems in General Practice of Medicine, South. M. J. **37**:90, 1944.

147. Plummer, N., and Wheeler, C.: Toxicity of Sulfadiazine: Observations on 1,357 Cases, Am. J. M. Sc. **207**:175, 1944.

148. Geever, E. F.: Pathologic Changes in Sulfadiazine Intoxication, Am. J. M. Sc. **207**:331, 1944.

149. Gessler, C. N.: Deaths from Sulfonamides; A Clinical and Pathological Study, with a Report of Three Cases, South. M. J. **37**:365, 1944.

blatt,[150] in which reference is made to the occurrence of areas of focal necrosis in the bone marrow, which were similar to the focal necrotic areas seen in other organs. The patients in these cases, however, did not have agranulocytosis. It is the opinion of Gessler that the lesions of focal necrosis, agranulocytosis and periarteritis nodosa may all be due to a combination of biochemical, pharmacologic and allergic reactions.

A general discussion is given of the favorable effects of sulfonamide compounds, and reference is made to the little understood mechanism of the toxicity of these drugs. It is stated that the toxic phenomena are related to the concentration of the sulfonamide compounds in the blood and the duration of treatment with them but that these phenomena may occur in the absence of excessive blood concentration or prolonged therapy. Hence, these are not the sole factors. To say that the toxic reactions are due to an idiosyncrasy is "begging the question," since so little is known relative to aberrant drug responses. In the opinion of the authors, sensitization appears to assume a role in some cases. They also emphasize the recent work indicating that the less absorbable sulfonamide compounds may cause vitamin deficiencies by decreasing the number of bacteria in the intestinal tract.

Hodges [151] reports observations on the prophylactic use of sulfadiazine against streptococcic and pneumococcic infections in an Army Air Force Technical School. One gram a day of sulfadiazine was given over a period of several weeks and in some instances 2 Gm. on two consecutive days once a week. He concludes that beneficial effects were attained against beta-hemolytic streptococcus infections, scarlet fever, streptococcic sore throat and pneumococcic infections. It is of interest that no serious untoward reactions occurred. No renal symptoms and no depression of the blood cells were reported.

Lee [152] reports the reactions following the massive administration of sulfadiazine to 25,000 men and women as a prophylactic against pneumococcic, streptococcic and meningococcic infections. A total of 2 Gm. of sulfadiazine was given. He reports that 0.50 per cent showed "reactions," 0.036 per cent showed "serious reactions" and 3 patients were "critically ill." No cases of agranulocytosis were observed. It is of interest to note that most of the seriously ill patients gave a history of previous administration of sulfonamide drugs, with reactions. While it is doubtful in our minds that the administration of a single dose of 2 Gm. of sulfadiazine would be productive of agranulocytosis, nevertheless it is a remote possibility which cannot be excluded in this report, as blood counts were not given. However, this clinical syndrome did not appear.

The comparative toxicity of sulfadiazine and sulfathiazole in children was studied by Dowrie and Abramson.[153] It is interesting to note that the commonest toxic effect was a fall in the polymorphonuclear cell count below 3,000 cells per cubic millimeter. This occurred in about 40 per cent of the patients in each series, with no great difference between the action of the two drugs. They note that this rate of occurrence of leukopenia is about ten times that reported in any other series, which leads them to believe that the incidence of granulocytopenia is much greater in children than in adults. It is their opinion that a fall in the polymorphonuclear cell count below normal levels does not necessarily indicate that agranulocytosis will develop, and no such complication was observed in their series. In patients for whom use of the drug was continued despite granulocytopenia, the blood counts rose slowly to normal. The authors caution, however, that except in case of the most serious infections the drug should be stopped when there are less than 1,000 polymorphonuclear cells per cubic millimeter. No correlation was discovered between the incidence of granulocytopenia and the daily dose of the drug, the total dose or the length of time of administration of the drug. They conclude that this toxic effect is due to obscure variable factors.

In studying the effects of succinylsulfathiazole on 45 patients, it has been found by Archer and Lehman [154] that the only unfavorable occurrence was a hemorrhagic cutaneous eruption which was seen in a patient who had previously received sulfathiazole. They state that the rash did not progress while the administration of the drug was continued and cleared up promptly when the drug was withheld. There were no changes in the blood picture in any of the patients.

150. Lederer, M., and Rosenblatt, P.: Death During Sulfathiazole Therapy, J. A. M. A. **119**:8 (May 2) 1942.

151. Hodges, R. G.: The Use of Sulfadiazine as a Prophylactic Against Respiratory Diseases, New England J. Med. **231**:817, 1944.

152. Lee, R. V.: Reactions Following Mass Administration of Sulfadiazine, J. A. M. A. **126**:630 (Nov. 4) 1944.

153. Dowrie, J. O., and Abramson, M. H.: Comparative Toxicities of Sulfadiazine and Sulfathiazole in Children, J. Pediat. **24**:176, 1944.

154. Archer, H. L., and Lehman, E. P.: Clinical and Laboratory Experiences with Succinylsulfathiazole, Ann. Surg. **119**:518, 1944.

Page [155] has reported on the treatment of 520 patients with acute bacillary dysentery with sulfaguanidine during a five week period ending June 28, 1943, in Northwest Africa. During this time the patients were given a ten day course of sulfaguanidine, averaging a total dose of 130 Gm. He reports that there were no deaths or serious complications and that close observation of the blood revealed no anemias or leukopenias.

The important practical point concerning the possibility that a person may be sensitive to a number of sulfonamide drugs is discussed by Park.[156] He reviews the literature which suggests that there may be either single or multiple sensitivity to such drugs when they are given internally or as determined by cutaneous tests. The author studied 40 cases of both internal and eczematous allergy by cutaneous tests and with oral medication with various sulfonamide drugs and related compounds, sulfanilic acid and procaine. He concluded that in 60 per cent of the cases the allergy was confined to one sulfonamide drug, whereas in 40 per cent it occurred to multiple sulfonamide drugs and to sulfanilic acid. He claims that the sensitivity of about half the patients who had multiple sensitivity was due to reaction to the sulfonamide radical ($NH_2.C_6H_4SO_2$). Of the other half it was to the aminophenyl radical ($NH_2C_6H_4.$), reactions also occurring to procaine. No cases were observed in which there was evidence of sensitization to still smaller groups than these.

A case of agranulocytosis following the administration of sulfapyridine is reported by Park, and a discussion of the mechanism responsible for the reduction in the neutrophils is given. After an examination of the reported cases of neutropenia from other drugs, Park states the opinion that all such cases are based on allergic responses. He also believes that other toxic effects of the sulfonamide drugs are probably on the same basis. It is his opinion that as a working rule chemotherapy should not be prolonged beyond the period of one week, since it may induce a dangerous form of drug allergy. Furthermore, most amenable infections respond within a few days, and longer administration is seldom necessary. It is his belief that sensitivity develops gradually, the first indications being a depression of the neutrophils in the blood. Following this there are constitutional symptoms, such as malaise and headache, which may go unnoticed in a patient already ill from the acute infection. With the complete disappearance of the neutrophils, the alarming syndrome of agranulocytosis suddenly appears. If sulfonamide therapy for longer than one week is necessary, then daily leukocyte counts should be made, and at the first sign of neutropenia the drug should be stopped and its elimination encouraged by promoting diuresis. He is unable to say how long this alleged allergic state persists, but it lasts perhaps for a period of several weeks if it is on the same basis as sulfonamide eruptions. Until this point is cleared up, however, he cautions that a victim of neutropenia requiring a sulfonamide compound subsequently should be given it in ascending doses with extreme caution.

A case of acute and fatal agranulocytosis with toxic dermal lesions produced by the therapeutic use of sulfathiazole in an infant 8 weeks old is reported by Kato, Sherman and Cannon.[157] At the time of this report no other cases of agranulocytosis in infants following sulfathiazole medication had been recorded, although instances of extreme granulocytopenia in children due to other members of the sulfonamide group had been observed. In this case, a total of 4.153 Gm. of sulfathiazole was given over a period of five weeks with frequent interruptions. At the end of this time the classic manifestations of agranulocytosis developed, which terminated fatally. These observations were confirmed by autopsy.

A case of a 22 year old white woman in whom agranulocytosis developed after she had received sulfadiazine and who recovered is reported by Meyer.[158] This patient received 44 Gm. of sulfathiazole as treatment for osteomyelitis of the terminal phalanx. The drug was discontinued because fever and a rash developed. The white blood cell count at this time was normal. Two days later the patient was given 1 Gm. of sulfadiazine every four hours, and after she had received 72 Gm. of this drug the white blood cell count was found to be 3,000 per cubic millimeter. A differential count showed 1 per cent segmented forms, 1 per cent stab forms, 3 per cent basophils and 95 per cent lymphocytes. The hemoglobin content was 85 per cent. The lesions of the mucous membranes characteristic of agranulocytosis appeared. Recovery followed the use of pentnucleotide, liver extract, blood transfusion and high intake of vitamin C.

155. Page, S. G.: Sulfaguanidine in Treatment of Acute Bacillary Dysentery: Study of Five Hundred and Twenty Cases, Virginia M. Monthly **70**:561, 1943.

156. Park, R. G.: Pathogenesis of Sulfonamide Neutropenia, Lancet **1**:401, 1944.

157. Kato, K.; Sherman, M. S., and Cannon, P. R.: Fatal Agranulocytosis Following Sulfathiazole Therapy: Report of Infant with Toxic Dermatosis, J. Pediat. **22**: 432, 1943.

158. Meyer, A. H.: Agranulocytosis: Report of Case Caused by Sulfadiazine, California & West. Med. **60**:277, 1944.

The case of an 18 year old youth in whom agranulocytosis developed after he had received 31 Gm. of sulfadiazine over a period of twenty-eight days because of an infection of the upper respiratory tract is reported by Blue.[159] The drug was discontinued when a sore throat, enlarged lymph nodes, cough and evidence of bronchopneumonia in the right side of the thorax appeared. His white blood cell count was found to be 1,950 per cubic millimeter, with 97 per cent lymphocytes and 3 per cent monocytes. Treatment was with repeated blood transfusions and pentnucleotide. The patient recovered, despite the fact that during the course of his illness he had acute catarrhal fever, lobar pneumonia, agranulocytic angina, acute myocarditis, acute otitis media, cervical adenitis and pyelonephritis.

The case of a patient 25 years of age in whom classic evidences of agranulocytosis developed after the administration of sulfadiazine is reported by Weinberg.[160] After receiving 102 Gm. of sulfadiazine orally for a period of twenty days, this patient had a complete absence of granulocytes in the circulating blood and the white blood cell count fell as low as 900 cells per cubic millimeter. The red blood cell count was not affected. With the discontinuance of the drug and administration of liver extract, pentnucleotide and multiple blood transfusions, the patient recovered from this complication but succumbed to subacute bacterial endocarditis, which was superimposed on rheumatic heart disease. It is of interest to note that after the patient recovered from agranulocytosis, sulfathiazole was given over a period of twenty-one days and aminopyrine for an interval of almost two months without a depression in the leukocyte count. Hettig and Sturgis have previously stressed the possible etiologic significance of a vitamin deficiency in agranulocytosis.[161] It is of interest to note that the patient had lost a total of 33 pounds (15 Kg.) in weight in four weeks just prior to the onset of the agranulocytosis. There is no indication in the report that the patient had a vitamin deficiency, but the loss of such a large amount of weight suggests this as a possibility.

The first case of agranulocytosis following the administration of sulfamerazine is added to the literature by Favorite, Reiner and London.[162] The patient was a white man 51 years of age who was given 42 Gm. of sulfamerazine over a period of twenty-three days for acute pharyngitis. The white blood cell count dropped as low as 600, and there was at times a total absence of all granulocytes. There were no important changes in the red blood cell count, hemoglobin content or platelets. The patient was treated with pentnucleotide and liver extract in large doses, but the authors properly conclude that it is impossible to say whether its administration influenced the course of the disease favorably. Improvement followed five days after these drugs were first given and six days after the discontinuance of sulfamerazine. As far as we know, no other cases of agranulocytosis have been reported as due to sulfamerazine.

The case of a man aged 42, in whom acute agranulocytosis developed thirteen days after a laparotomy for gastric ulcer was performed, is reported.[163] At the time of the operation 5 Gm. of sulfanilamide crystals was scattered along the suture lines and over the area of resection because of presumed contamination. Thirteen days later the white blood cell count was 2,700 per cubic millimeter and there was a decided increase in the number of nonsegmented forms of neutrophilic leukocytes. After an interval of four more days, that is, seventeen days after the operation, the granulocytes had almost disappeared, and they remained noticeably decreased until the patient's death, nineteen days after the operation. The patient received no additional sulfanilamide or any other drugs which might have caused the condition. So far as we are aware, this is the first reported instance in which agranulocytosis has been observed to follow the local or topical application of one of the sulfonamide drugs.

The first case in the literature, as far as we know, of agranulocytosis following the use of sulfaguanidine is reported by Grant.[164] The patient was a 15 year old girl, who was given the drug for the treatment of Shigella paradysenteriae Sonne infection. After she received 25.2 Gm., with resultant improvement, the therapy was discontinued for an interval of six days and then resumed on account of a recurrence of symptoms until 15 Gm. more had been admin-

159. Blue, J. A.: Allergic Agranulocytosis with Complications: Case Report, Am. J. M. Sc. **207**:453, 1944.

160. Weinberg, H. B.: Agranulocytosis Following Sulfadiazine Administration, J. Iowa M. Soc. **34**:63, 1944.

161. Hettig, R. A., and Sturgis, C. C.: Sulfadiazine Agranulocytosis, J. Michigan M. Soc. **42**:959, 1944.

162. Favorite, G. O.; Reiner, L., and London, R.: Acute Agranulocytosis During Sulfamerazine Therapy, J. Lab. & Clin. Med. **29**:899, 1944.

163. Arrowsmith, W. R.; Binkley, B., and Moore, C. V.: Fatal Agranulocytosis Following the Intraperitoneal Implantation of Sulfanilamide Crystals, Ann. Int. Med. **21**:323, 1944.

164. Grant, A.: Severe Granulocytopenia from Sulphaguanidine, Brit. M. J. **1**:557, 1944.

istered. At this time it was discontinued on account of a febrile response. The following day the white blood cell count was found to be 4,100 per cubic millimeter, with 38 per cent neutrophils; a day later the count was 2,000 per cubic millimeter, with only 10 per cent neutrophils. The patient recovered after the discontinuance of the drug and the use of pentnucleotide. The author states that the Medical Research Council Memorandum 1943 b emphasizes the fact that two thirds of the amount of sulfaguanidine administered orally is absorbed before the ileocecal valve is reached. He cites Firor and Poth [165] as stating that the relatively low concentrations of this drug in the blood are due in far greater measure to rapid elimination in the urine than to poor absorption from the intestine. He considers, therefore, that the possibility of agranulocytosis resulting from the administration of sulfaguanidine merits attention.

It is stated by Sherlock and White [166] that the literature contains records of only 18 cases of purpura in association with sulfonamide administration. They give the details of a case as follows. A man aged 52 was treated with sulfapyridine for pneumonia. He received 30 Gm. in seven days, after which his temperature fell to normal and the drug was discontinued. On account of a second rise in temperature, he was given 4.5 Gm. during the next five days. Fever appeared after each dose of sulfapyridine. Extensive evidences of purpura developed, with bleeding from the nose and gums and the vomiting of blood as well as with a purpuric eruption. The hemoglobin content was 47 per cent; the red blood cell count was 2,500,000 per cubic millimeter, leukocytes 10,000 per cubic millimeter, platelets 150,000 per cubic millimeter, bleeding time thirty-six and one-half minutes, coagulation time twenty-seven minutes and clot retraction in one hour was 19 per cent, whereas the normal is regarded as 44 to 66 per cent. The capillary resistance test by positive pressure method indicated a decided increase in fragility. Sternal marrow showed that megakaryocytes were present (0.4 per cent), and these appeared normal. The patient died, and just before death it was observed that his blood pressure was 260 systolic and 140 diastolic and that the plasma urea content was 214 mg. per hundred cubic centimeters. It was considered that the recent hemorrhagic changes in the kidney were responsible for the impairment of renal function. In addition the authors point out that gastrointestinal hemorrhage could play a part in producing azotemia. It was concluded that the hemorrhagic state in their patient was, therefore, probably the cause rather than the result of the uremia. They emphasize the importance of the capillaries in the pathogenesis of the purpura and state that in the 7 recorded cases of purpura following sulfonamide therapy in which the resistance of the capillaries was tested an extreme fragility was noted. In 4 of the reported cases, including their own, the platelet count, although low, had not been reduced to the level usually associated with bleeding in essential thrombopenic purpura. They state that in the reported cases no member of the sulfonamide group of drugs seemed exempt as a causative agent, and sulfanilamide, sulfapyridine, sulfathiazole and sulfadiazine have all been held responsible. The high mortality of this condition, as indicated by a fatal termination in 8 of the 9 cases, makes early recognition essential. In their opinion the first clinical sign is nearly always epistaxis. They believe that adequate transfusion of fresh blood is probably the best type of treatment.

The literature in reference to the occurrence of thrombopenic purpura following sulfonamide therapy is reviewed briefly by Brillinger,[167] and a case report is presented. At the time the article was written he stated that there were 18 cases of thrombopenic purpura following the administration of various sulfonamide drugs reported in the literature. These include cases of purpura following the use of sulfanilamide, azosulfamide, sulfapyridine, sulfathiazole and sulfadiazine. He emphasized particularly that in all but 3 of these cases the purpuric reaction occurred while the drug was being given or at least on the day after it was discontinued. The longest interval between the cessation of use of the drug and the appearance of the purpura was eleven days. Two of the three delayed reactions followed administration of sulfathiazole. His own patient was a healthy man 29 years of age who previously had not been ill. He was treated with 360 grains (23.3 Gm.) of sulfathiazole over a period of five days, at which time the use of sulfathiazole was discontinued because of albuminuria with many sulfonamide crystals in the urine. The purpura appeared for the first time on the eighteenth day, some thirteen days after the cessation of the medication. This was accompanied with a high leukocyte count and by a decidedly leukemoid

165. Firor, W. M., and Poth, E. J.: Intestinal Antisepsis, with Special Reference to Sulfanilylguanidine, Ann. Surg. **114**:663, 1941.

166. Sherlock, S., and White, J. C.: A Fatal Case of Purpura After Sulphapyridine, Brit. M. J. **2**:401, 1944.

167. Brillinger, H. R.: A Case of Delayed Thrombopenic Purpura Following Sulfathiazole Therapy, J. Canada. M. Serv. **1**:324, 1944.

blood picture. It is stated that the lowest platelet count was 110,000 per cubic millimeter; the bleeding time was three minutes and the clotting time eleven minutes. The patient recovered completely after two small transfusions of citrated blood.

Meyer [168] states that in the literature there are 2 previous reports of thrombopenic purpura developing from the use of sulfadiazine. These are reported by Kracke and Townsend [169] and by Gorham and others.[170] The patient reported on by Meyer was a 7 year old boy who became ill with chickenpox and in whom impetiginous lesions subsequently developed. Sulfathiazole ointment for local application was prescribed, and sulfadiazine was given amounting to a total of 4:5 Gm. in one and one-half days, at which time purpuric spots were noted about the knees and buttocks. An additional 3 Gm. was administered in two and one-half days, at the end of which time the urine became bloody; after a total of 9 Gm. had been given, the hematuria became so pronounced that the child was hospitalized. There were purpuric spots over the entire body. The hemoglobin content was 23 per cent with 1,433,000 red blood cells per cubic millimeter, platelets 20,000 per cubic millimeter and white blood cells 23,850 per cubic millimeter, with 85 per cent segmented neutrophils. Bleeding time was forty-five minutes and the clotting time six minutes, with little retraction after eighteen hours. The reaction to the tourniquet test was strongly positive. The urine was grossly bloody. The patient was given transfusions with a total of 1,750 cc. of blood in units of 300 cc. Two weeks after his admission to the hospital the bleeding time was one and one-half minutes, the platelet count 139,000 per cubic millimeter, the hemoglobin content 89 per cent and the urine normal. The patient made a complete recovery.

Reference is made by Latven and Welch [171] to the previous work of Richardson,[172] who reported that in mice hemolytic anemia of variable intensity develops within a period of two weeks when sulfanilamide, sulfaguanidine, sulfapyridine or sulfathiazole is incorporated in a stock ration. The potentiality of each of these sulfonamide compounds for the production of hemolytic anemia was related by Richardson to the concentration of the compound within the erythrocytes. Latven and Welch have studied the hemolytic anemia–producing properties of sulfapyridine, sulfamerazine and sulfadiazine, which have been evaluated in mice. This evaluation has been accomplished by determining the percentage incidence of anemia produced by various concentrations of sulfonamide compounds in the blood. They found that the concentration of sulfonamide compounds in the blood necessary to produce a 50 per cent incidence of anemia in mice was as follows: with sulfapyridine 2.8 mg., with sulfadiazine 33 mg. and with sulfamerazine 31 mg. per hundred cubic centimeters. They conclude that a large number of cases will be required to prove conclusively that in human beings, as in mice, sulfadiazine is decidedly less productive of hemolytic anemia than are sulfathiazole and sulfapyridine.

A case of acute hemolytic anemia following the administration of 39 Gm. of sulfadiazine to a patient with bronchopneumonia for a period of nine days is reported by Layne and Schemm.[173] The anemia in this patient became very severe, as the hemoglobin content was 3.75 Gm. per hundred cubic centimeters (24 per cent) and the red blood cell count 1,270,000 per cubic millimeter. The authors point out that improvement in the patient's anemia occurred coincidentally with the administration of 90 units of liver extract on two successive days. They recognize that this may have played no role in the patient's recovery. Some evidence has been offered, however, to suggest that liver extract is effective in correcting the granulocytopenia and the anemia in rats fed various sulfonamide drugs. They conclude that this form of therapy may have contributed to the patient's recovery.

AGRANULOCYTOSIS DUE TO VARIOUS CAUSES

Thiouracil.—In a comprehensive article on the use of thiouracil in the treatment of hyper-

168. Meyer, A. H.: Thrombocytopenic Purpura: Case Caused by Sulfadiazine, California & West. Med. **60**:98, 1944.

169. Kracke, R. R., and Townsend, E. W.: The Effect of Sulfonamide Drugs on the Blood Platelets: Report of Two Cases of Thrombopenic Purpura and Experimental Studies on Patients Receiving Sulfonamide Drugs, J. A. M. A. **122**:168 (May 15) 1943.

170. Gorham, L. W.; Propp, S.; Schwind, J. L., and Climenko, D. R.: Thrombocytopenic Purpura Caused by Sulfonamide Drugs: A Report of Three Cases, Am. J. M. Sc. **205**:246, 1943.

171. Latven, A. R., and Welch, A. D.: Sulfamerazine (2-Sulfanilamido-4-Methylpyrimidine): III. Comparative Activity of Sulfamerazine, Sulfadiazine and Sulfapyridine in Production of Hemolytic Anemia in Mouse, J. Pharmacol. & Exper. Therap. **81**:301, 1944.

172. Richardson, A. P.: Comparative Effects of Sulfonamide Compounds as to Anemia and Cyanosis, J. Pharmacol. & Exper. Therap. **72**:99, 1941.

173. Layne, J. A., and Schemm, F. R.: Acute Macrocytic Hemolytic Anemia Occurring Following Administration of Sulfadiazine, J. Lab. & Clin. Med. **29**:347, 1944.

thyroidism, Astwood[174] discusses the various toxic complications which may develop during the course of the therapy. According to him the most serious one is agranulocytosis, and he reports 2 cases in detail. In 1 the patient was given the excessive dose of 2 Gm. daily and recovered after the omission of the drug. It is of interest that the same patient subsequently experienced a febrile reaction to sulfathiazole given while he had a scarlatiniform eruption following the administration of phenobarbital. Astwood suggests, therefore, the possibility that persons in whom allergic reactions readily develop are more likely to become sensitized to thiouracil. Another patient, who had received 0.6 Gm. of thiouracil for sixty-two days and 0.3 Gm. for an additional thirty-one days, had a streptococcic infection of the finger on the eighty-fifth day but did not return to the clinic for a week. In that interval she had received 4 Gm. of sulfamerazine daily for three days and 3 Gm. daily thereafter. When first determined, the total white blood cell count was 2,000, of which 5 per cent were granulocytes. Discontinuance of both the thiouracil and the sulfonamide therapy, with local treatment of the digital infection and of the ulcerative stomatitis and pharyngitis, was followed by complete recovery. The differential count became normal within ten days. It is impossible in this case to determine to what degree thiouracil, sulfamerazine and the infection may each have contributed to the leukopenia. Astwood also states that had the patient been under more careful observation the complication might have been detected before reaching an alarming degree of severity. A number of instances of leukopenia of varying degree occurred in the cases in his series. Although the total leukocyte count fell to 3,000 or 4,000 per cubic millimeter or less on some occasions, it subsequently rose in spite of the fact that the drug was continued. In Astwood's opinion "it is uncertain" whether or not all these mild leukopenias are due to the drug. He states that about 10 per cent of all patients who receive thiouracil will have toxic manifestations, either febrile or in association with a leukopenia. The best indications of impending danger of toxic reactions, he believes, are the subjective sensations of the patient and the body temperature. Frequent leukocyte counts, while helpful, apparently are not as reliable as clinical symptoms in indicating the early onset of serious complications. Serious side effects can probably be avoided if the patients are instructed to seek medical advice as soon as abnormal symptoms are experienced.

Williams and Clute[175] reported the effect of thiouracil on 72 patients, including a continuation of the study of 9 patients previously reported by Williams and Bissell.[176] The most serious complication encountered was leukopenia with agranulocytosis, which occurred in 1 patient. This occurred at the end of six weeks of treatment, during which time the patient had received 0.6 Gm. of thiouracil daily for the first two weeks, 0.4 Gm. daily for the next two weeks and 0.2 Gm. daily for the last two weeks. The leukocyte count was determined when the patient complained of an infected finger and fever. Treatment consisted of pentnucleotide and large doses of crude liver extract. Complete recovery occurred, although for a period of seven days only a few granulocytes were present in the circulating blood. In another case the white blood cell count dropped to 3,200 with 20 per cent granulocytes only a few days after treatment with thiouracil was begun. Despite continued therapy the white blood cell count returned to normal and the granulocytes rose 50 per cent. Subsequently the patient took thiouracil for more than six months and remained in good health. Leukopenia and granulocytopenia were seen in some other cases, but the incidence was apparently no greater than that observed in untreated thyrotoxic patients. These authors conclude that of the various complications encountered thus far the only one to cause appreciable concern is agranulocytosis.

In the review of the action of antithyroid drugs, with particular reference to thiouracil, Williams[177] has this to say in regard to the toxic actions of thiouracil. According to him, such untoward effects are to be anticipated in about 10 per cent of the patients; most of the complications have appeared in the first five weeks of treatment. These have consisted of fever, morbilliform rash, urticaria, arthritis, vomiting, diarrhea, enlargement of the submaxillary salivary glands, edema of the legs, lymphadenopathy, leukopenia and agranulocytosis. In his opinion, the last of these is the only serious toxic reaction. At the time his article was written only 2 cases of toxic reaction to thiouracil with death due to agranulocytosis had been reported, but Williams states that he has knowl-

174. Astwood, E. B.: Thiouracil Treatment in Hyperthyroidism, J. Clin. Endocrinol. **4**:229, 1944.

175. Williams, R. H., and Clute, H. M.: Thiouracil in the Treatment of Thyrotoxicosis, New England J. Med. **230**:657, 1944.

176. Williams, R. H., and Bissell, G. W.: Thiouracil in the Treatment of Thyrotoxicosis, New England J. Med. **229**:97, 1943.

177. Williams, R. H.: Antithyroid Drugs, with Particular Reference to Thiouracil, Arch. Int. Med. **74**:479 (Dec.) 1944.

edge of several other cases which have not been published. Of 170 patients he has treated, 2 have had agranulocytosis. Undoubtedly other patients do not have the outspoken syndrome of agranulocytosis but do have an important leukopenia, which is an indication that the patient's treatment should be stopped or continued under careful supervision. According to Williams, except for fever, arthritis and leukopenia, the complications have been observed to disappear when the dosage of thiouracil was reduced.

The case of a 44 year old woman with exophthalmic goiter of one year's duration who received thiourea and in whom purpura developed is reported by Newcomb and Deane.[178] Thiourea in a dosage of 1 Gm. three times a day was given for three weeks. Ten days after the use of the drug had been started, the signs of hyperthyroidism began to recede, at which time the dosage of thiourea was reduced to 1 Gm. twice a day for a week and then 1 Gm. daily. Two weeks after the drug had been started, the leukocyte count was 9,200 per cubic millimeter, with 70 per cent granulocytes and a normal differential count. After a total of 83 Gm. had been given, namely, five weeks after the beginning of treatment, epistaxis and a generalized purpura developed, with ecchymoses and purpuric spots on the buccal mucous membrane. The administration of thiourea was immediately stopped. The following day the ecchymoses became more numerous, and there was bleeding from the gums. There were a normal red blood cell count and hemoglobin content. The white blood cell count was 4,100 per cubic millimeter with 24 per cent granulocytes; the platelets were 18,000 per cubic millimeter. The bleeding time was greatly prolonged, and at the end of four hours bleeding from the venipuncture wound showed no signs of ceasing. The coagulation time was normal. Sternal puncture revealed no important changes except that the platelets were scanty and only 2 megakaryocytes were seen. The white blood cell count fell to 3,700 per cubic millimeter, with 16 per cent polymorphonuclear neutrophils, and there were 9,000 platelets per cubic millimeter. She was given 500 cc. of fresh whole blood, and all bleeding promptly ceased. A few doses of pentnucleotide were given. The patient made a complete recovery, and twelve days after the appearance of purpura the leukocyte count was 6,200, with 36 per cent granulocytes and 67,000 platelets. The report of this case is important because not only granulocytopenia but also thrombopenic purpura developed.

McGavack and his associates[179] reviewed the toxic reactions due to thiouracil in 135 patients with toxic goiter. In this group 2 had agranulocytosis and recovered. In addition, a transient leukopenia appeared in 3 patients. One of the patients in whom agranulocytosis developed was a 50 year old white woman who received 0.5 Gm. of thiouracil daily for forty-one days. At this time fever, sore throat, gingivitis, headaches and enlargement and tenderness of the cervical lymph nodes developed. The blood count taken at this time showed 3,500 white blood cells per cubic millimeter, with 97 per cent lymphocytes. With cessation of the use of the drug and with the administration of several blood transfusions, recovery was complete. In their opinion it would appear that omission of the drug without any recourse to transfusions would have adequately relieved her condition. In 3 of the patients observed by this group there were decreases in the white blood cells to 3,500, 4,500 and 2,500 per cubic millimeter, and in the percentage of polymorphonuclear leukocytes to 41, 32 and 48, respectively. The blood counts returned to normal without a change in the treatment of any of these 3 patients. It is questionable, therefore, in their opinion whether one should regard these responses as pathologic or as physiologic, and it is difficult to be sure whether leukopenia with lymphocytosis can be considered as a toxic phenomenon. In any event, it hardly warrants complete cessation of therapy. A more satisfactory course would be to consider such responses as an indication for temporary or permanent decrease in the size of the dose employed. From our experience, it would seem that leukopenia occurs more commonly in patients who are receiving thiouracil than would be indicated by the reports in the literature. In our group of patients for whom leukocyte counts were determined every other day, a moderate leukopenia without evidence of agranulocytosis occurred in a great percentage of patients. In almost all instances, however, with continuation of the therapy or its interruption for only one or two days, there was a return of the white blood cell count to normal. Until knowledge is more complete on this subject, however, any significant decrease in the white blood cell count with a diminution in the number of polymorphonuclear leuko-

178. Newcomb, P. B., and Deane, E. W.: Thiourea Causing Granulopenia and Thrombopenia, Lancet **1**:179, 1944.

179. McGavack, T. H.; Gerl, A. J.; Vogel, M., and Schwimmer, D.: Treatment of Twenty-Six Thyrotoxic Patients with Thiouracil and Review of Toxic Reactions in All (135) Reported Cases, J. Clin. Endocrinol. **4**:249, 1944.

cytes is an indication to observe the patient with extreme care.

It is reported by Kahn and Stock [180] that as far as can be determined their case is the first in which death occurred from agranulocytosis attributable to thiouracil. The patient was a 62 year old woman with severe and uncontrolled diabetes and a diffuse toxic goiter, with a basal metabolic rate of + 65 per cent. The diabetes was well controlled, and the patient was given thiouracil, 0.6 Gm. daily for forty-four days and then 0.4 Gm. for eight days. A total dose of 30.8 Gm. was given in fifty-four days. On the fifty-first day of treatment, the body temperature was elevated to 101.4 F. and the patient complained of a sore throat. Within twenty-four hours the white blood cell count was found to be 1,100 per cubic millimeter, with 2 per cent granulocytes. The patient died five days after the onset of the agranulocytosis, despite all forms of therapy, including blood transfusions, use of crude liver extract, pentose nucleotide, extract of yellow bone marrow and penicillin. It was assumed that the thiouracil was the cause of the toxic hepatitis which was also present as well as of the agranulocytosis. The severe diabetes mellitus was regarded as a contributory factor in the death of the patient.

David Haller states,[181] "Thiouracil is a dangerous drug, and at least half a dozen fatal cases of agranulocytosis have been described following its use [no reference or further details are given], and only this week I conducted a necropsy upon such a case, the inquest resulting in a verdict of misadventure following the administration of thiouracil." In the author's opinion, the drug should be given only under the strictest control in a hospital.

In a general article on the use of thiouracil in the preparation of patients with hyperthyroidism for thyroidectomy, Moore and his associates [182] state that in their opinion thiouracil exhibits a toxicity similar to that of the sulfonamide drugs. Among 52 patients treated up to the time the article was written, they observed 2 with leukopenia, 4 with oral infections possibly related to the medication and 1 with a generalized lymphadenopathy which cleared after discontinuance of the drug. In the patients with leukopenia the white blood cell count was about 2,500 per cubic millimeter. The polymorphonuclear leukocytes were never below 30 per cent. In treatment of 1 patient the drug was continued until the count was normal; in treatment of the other the use of the drug was stopped. In no case, however, in the experience of these observers, has any toxic manifestation either prevented or complicated operative removal of the gland.

St. Johnston [183] reports on 3 patients with toxic goiter treated with thiourea in doses of 3 Gm. daily for periods of seven to eight days who had temperatures of 100.4 to 101 F., with palpable enlargement of the spleen, a fall in the white blood cell count with monocytosis and a maculopapular eruption. There was no pronounced drop in the white blood cell count in any instance, the lowest being 4,600 white blood cells per cubic millimeter, with 51 per cent polymorphonuclear leukocytes. The enlargement of the spleen was unexplained.

Rubinstein [184] reports the case of a woman 47 years old, who suffered from carcinoma of the thyroid gland. She was given thiouracil in a dosage of 0.2 Gm. four times a day for approximately fifteen weeks. It is of interest to note that the thyroid nodes became somewhat diminished in size and softer in the supraclavicular region. After a period of fifteen weeks the white blood cell count was observed to be 1,700 per cubic millimeter and a few days later it fell to 750. At this time there were 4 per cent myelocytes and 10 per cent neutrophils. Studies of the bone marrow showed an arrested maturation associated with hypoplasia of the myeloid elements of the bone marrow, which was taken as the explanation for agranulocytosis of the peripheral blood. After the use of the drug was discontinued, a complete hematologic recovery followed. A return to normal of the bone marrow preceded that of the peripheral blood.

A patient observed by Linsell [185] received continuous thiouracil therapy for four months, with a daily dose of 1 Gm. for one month and 0.2 Gm. daily subsequently. The neutropenia developed after ten weeks of treatment, when the patient had taken approximately 44 Gm. At this time she had an acute febrile attack with some ulceration of the mouth and sore throat, which cleared up entirely without treatment within six days. After twelve weeks of addi-

180. Kahn, J., and Stock, R. P.: Fatal Agranulocytosis Resulting from Thiouracil, J. A. M. A. **126**:358 (Oct. 7) 1944.

181. Haller, D.: Letter to Brit. M. J. **1**:382, 1944.

182. Moore, F. D.; Sweeney, D. N., Jr.; Cope, O.; Rawson, R. W., and Means, J. H.: Use of Thiouracil in Preparation of Patients with Hyperthyroidism for Thyroidectomy, Ann. Surg. **120**:152, 1944.

183. St. Johnston, C. R.: Toxic Reaction to Thiourea: Report on Three Cases, Lancet **2**:42, 1944.

184. Rubinstein, M. A.: Agranulocytosis Following Thiouracil Administration, Am. J. Clin. Path. **14**:540, 1944.

185. Linsell, D.: Agranulocytosis After Use of Thiouracil, Brit. M. J. **2**:597, 1944.

tional treatment with thiouracil during which time she took 48 Gm., she had a much severer febrile attack and was admitted to the hospital for observation. The classic signs of agranulocytosis were present, and, in addition, there was a severe conjunctivitis of the left eye with cellulitis spreading into the orbital tissue. At this time the patient had a white blood cell count of 1,200 per cubic millimeter, and "an occasional degenerate polymorphonuclear cell" was seen in the peripheral blood. Recovery was rapid after the administration of thiouracil was discontinued, although the author attributes some improvement to repeated transfusions of fresh blood. In addition, the patient received pentnucleotide and ascorbic acid.

It is reported by Welshman [186] that thiouracil had been employed in the treatment of 2 patients with thyrotoxicosis and "in both the total white cell count fell to about 2,200 per cubic millimeter with depression of the polymorphs." With discontinuance of the use of the drug, the white cell count rose to 10,000 in five days. Thyroidectomy was performed, and both patients made an uneventful recovery. It was suggested that the drug should be employed with caution and should be used only when there are facilities for making frequent white cell counts.

A study was made of the various tissues of rats after they had received a relatively large amount of thiouracil. Microscopic examination of the bone marrow of approximately 50 rats revealed no definite abnormalities. They had been treated for from seven to ninety days. A few studies were made of the circulating blood obtained from the heart. In several instances the white blood cell count was low (the lowest was 3,600 per cubic millimeter), but the average was 9,000 per cubic millimeter.

Favorable results are reported by Reveno [187] in the treatment of 9 patients. For 6 the results were good, for 3 they were not so satisfactory for various reasons, such as lack of cooperation. No instance of agranulocytosis or anemia occurred.

In treating 11 patients with severe hyperthyroidism with 0.6 Gm. of thiouracil daily, Bartels [188] did not observe any toxic effects from the drug. According to him, the white blood cell counts were carefully observed during the course of treatment but no change occurred.

Gabrilove and Kert [189] report the treatment of 9 patients with thiouracil and observed that 3 exhibited manifestations of sensitivity to the drug. In 1 there were a febrile rise and a moderate generalized lymphadenopathy, suggesting that the patient had infectious mononucleosis, but there were a normal hemogram and a negative heterophile reaction of the blood. The second patient exhibited fever and dermatitis. In the third patient, a woman 47 years of age, leukopenia developed with a white blood cell count of 2,650 per cubic millimeter and a differential count of 59 per cent segmented polymorphonuclear neutrophils, 4 per cent nonsegmented polymorphonuclear leukocytes, 28 per cent lymphocytes, 4 per cent monocytes and 5 per cent eosinophils. This occurred after the patient had been given 1 Gm. of thiouracil daily in five divided doses for a period of sixteen days. The use of the drug was stopped, and the patient was then prepared for subtotal thyroidectomy by administration of iodine. She recovered without further complications.

Paschkis and his associates [190] found no toxic manifestations in the blood of 6 patients with thyrotoxicosis who received thiourea. Among the 15 patients with thyrotoxicosis who received thiouracil, toxic manifestations, including leukopenia (2,500 cells per cubic millimeter) developed in 1. This condition became apparent after an infection of the upper respiratory tract which developed after eight months of treatment. The leukocyte count returned to normal, and the rash disappeared three days after the drug was discontinued. Forty-eight hours after treatment was resumed, the leukocyte count fell again but the rash did not reappear. This patient was given 1 Gm. of thiouracil for the most part but at times received as much as 2 Gm. a day.

The case of a Negro man aged 40, with exophthalmic goiter, in whom agranulocytosis developed after the use of thiouracil, is reported by Meyer.[191] After the patient had received a total of 40.8 Gm. of thiouracil in fifty-five days in doses of 1 to 2 Gm., there were a severe chill, a sore throat and a rise in temperature to 104 F. The white blood cell count was 2,000 per cubic millimeter, with 19 per cent segmented neutrophils, 3 per cent juvenile forms and 71 per cent lymphocytes. Recovery followed

186. Welshman, B. C.: Effect of Thiouracil on the White Cells, Letter to the Editor, Lancet **1**:195, 1944.

187. Reveno, W. S.: Thyrotoxicosis Treated with Thiouracil, J. A. M. A. **126**:153 (Sept. 16) 1944.

188. Bartels, E. C.: Thiouracil: Its Use in the Preoperative Management of Severe Hyperthyroidism; Preliminary Report, J. A. M. A. **125**:24 (May 6) 1944.

189. Gabrilove, J. L., and Kert, M. J.: Sensitivity to Thiouracil: Report of Three Cases, J. A. M. A. **124**: 504 (Feb. 19) 1944.

190. Paschkis, K. E.; Cantarow, A.; Rakoff, A. E.; Walkling, A. A., and Tourish, W. J.: Thiourea and Thiouracil in Treatment of Thyrotoxicosis, J. Clin. Endocrinol. **4**:179, 1944.

191. Meyer, A. H.: Granulocytopenia: A Case Caused by Thiouracil, California & West. Med. **61**:54, 1944.

the discontinuance of the use of thiouracil and the administration of pentnucleotide and liver extract.

Miscellaneous Agents.—A review of the literature and general clinical observations on agranulocytic angina is given by Copley.[192] In this he reports that in the state of Virginia in the past eight years there have been 36 cases of agranulocytic angina which have been reported. There were two peaks in these years, one being in 1937, when there were 12 cases, and one in 1940, when there were 10. Three of the 18 deaths since 1930 were due to one of the sulfonamide drugs, according to replies sent by physicians reporting these deaths. One fatal case followed the administration of 108 Gm. of sulfadiazine over a period of twenty days. One patient, a man aged 54, in our opinion did not have agranulocytic angina. The patient had taken aminopyrine eleven months before the acute onset, which was preceded by a prodromal period of ill health. He was treated with the recommended therapeutic measures, namely, pentnucleotide, blood transfusions and liver extract administered parenterally, and lived three years and eight months. It is much more likely, in our opinion, that this patient died of subleukemic leukemia.

A comprehensive review on the subject of agranulocytosis and neutropenia is presented by Rodríguez.[193]

The rarity of blood dyscrasias following injections of the arsphenamine drugs is emphasized by Ferguson.[194] He states that in the United States Navy over a million injections of such drugs were given between the years 1925 and 1938 with only 24 resultant blood dyscrasias, which gives an incidence of roughly 1 in 50,000. He reports on 6 patients with hematologic disorders after treatment with neoarsphenamine. For the first 5 a diagnosis of agranulocytosis was made because the changes were largely limited to a reduction or complete disappearance of the granulocytes in the circulating blood. In the sixth patient, however, there was a moderate reduction in the red blood cells, to 4,100,000 per cubic millimeter, with a hemoglobin content of 72 per cent. The total white blood cell count was 1,200 per cubic millimeter, with 4 per cent polymorphonuclear leukocytes, and the blood platelets were 33,600 per cubic millimeter. Ferguson classified the illness of the sixth patient as acute aplastic anemia because all elements of the bone marrow were affected. The first 5 patients reported on, that is, those with agranulocytosis, received an average of five injections of arsenic with an average dose of 2.1 Gm. In the illness classified as aplastic anemia, the damage to the bone marrow was regarded as more extensive, as postmortem studies showed that the megakaryocytes and the precursors of the red blood cells were affected. In this patient symptoms did not begin until six or seven weeks after the beginning of antisyphilitic treatment. The author believes that in classifying the blood dyscrasias following arsphenamine simple thrombopenia should be grouped separately from the other dyscrasias which are due to involvement of the bone marrow. It is thought by this observer that an increased absolute number of monocytes, as found in 3 of the patients with agranulocytosis who recovered, was a favorable prognostic sign. This has previously been reported, but there is some difference of opinion as to how reliable it is from the standpoint of prognosis. In the cases of agranulocytosis, examination of the bone marrow by sternal puncture and at necropsy showed evidence of arrest of maturation similar to that found in cases of so-called idiopathic agranulocytosis. In the opinion of this observer pentnucleotide therapy "appears" to be effective in treatment of agranulocytosis provided treatment is not too long delayed.

Two cases of agranulocytosis are reported by Lana Martínez.[195] The first is that of a man 28 years old, who was treated for syphilis for about two years with arsphenamine, bismuth and mercury. Fever, malaise and increasing fatigue developed, at which time the blood was examined and the white blood cell count was found to be 5,300 per cubic millimeter, with no neutrophils. There were 25 per cent lymphocytes present and 75 per cent monocytes. The antisyphilitic medication was discontinued, and ascorbic acid was administered. Slow improvement followed, and one month later the blood was normal. The second case is that of a man 26 years old with syphilis and a gonococcic infection of the urethra. He was treated with arsphenamine and sulfonamide drugs, the latter in doses of 3 Gm. daily. After a few days fever, chills and malaise developed. Examination of the blood showed the white blood cell count

192. Copley, E. L.: Agranulocytic Angina—a Drug Hazard, Virginia M. Monthly **71**:416, 1944.

193. Rodríguez y Rodríguez, A.: Sobre la agranulocitosis o neutropenia, Medicina, Madrid (pt. II) **12**:491, 1944.

194. Ferguson, J. W.: Agranulocytosis and Aplastic Anemia After Arsphenamines: Report of Six Cases, Lancet **1**:334, 1944.

195. Lana Martínez, F.: Dos casos de agranulocitosis, Med. españ. **4**:383, 1940.

to be 4,600 per cubic millimeter, with no neutrophils. With the discontinuance of the medication and administration of ascorbic acid the patient improved, and one week later the blood was normal. In neither of the cases were there lesions of the mouth or throat.

A case was reported by Oppikofer [196] of a man 33 years old in whom the classic picture of agranulocytosis developed after the use of several drugs, including aminopyrine. The white blood cell count fell to 5,050 per cubic millimeter, with no granulocytes present in the peripheral blood. The patient was treated with "Nucleotrat" intramuscularly, 10 cc. twice daily. He was also given "a number" of blood transfusions. Recovery followed. A brief discussion of the clinical picture of agranulocytosis is given.

A case is reported by Estol Baleztena [197] of a child 10 years of age who had previously suffered from pulmonary tuberculosis. Two injections a week of a gold preparation (*Sonocrisina*) were given in a dose of 0.025 Gm. for each injection. After five injections, fever, sore throat and agranulocytosis developed. Recovery followed symptomatic treatment.

A case of agranulocytosis developing in a woman aged 45, in whom the white blood cell count fell to a minimum of 900 per cubic millimeter, with 10 per cent granulocytes, is reported by Rawls.[198] This patient had rheumatoid arthritis and had taken acetylsalicylic acid, acetophenetidin and neocinchophen at various intervals without any signs of toxicity. Agranulocytosis developed after she was said to have taken 7½ grains (0.48 Gm.) of cinchophen three times a day and 1 mg. of menadione (2-methyl-1, 4 naphthoquinone, synthetic vitamin K) three times a day for six days. It is "assumed" by the author that the synthetic vitamin K radical may have been responsible for this condition, as it contains the quinone radical, which, in his opinion, might produce agranulocytosis. Certainly the evidence is not clear in this case that the agranulocytosis was due to this particular drug.

A familial type of malignant chronic granulocytopenia is described by Béguez César.[199] This condition is characterized by albinism, indeterminate feverish states, nystagmus, leukopenia, granulocytopenia and lymphomonocytosis. The author considers it to be a familial disease. According to him it has the characteristics of a recessive mendelian trait. Of 13 brothers studied by him, 4 suffered from the condition. He concludes that of the 9 living brothers a number will transmit the disease while others will not.

Experiments were performed by Staub and Bucher [200] in which they injected solutions of glycogen into rabbits and concluded that the leukopenia which resulted was proportionate to the size of the glycogen molecules. When a large molecule solution was injected the leukopenia was greater than that produced by one containing small molecules. They conclude that leukopenia is dependent to a certain extent on the physical state of the molecules of the injected substance.

The case of a woman aged 26 is reported by Barling [201] in which the unusual feature was that for twelve years the patient had complained of ulcers in the mouth recurring at regular intervals of three to four weeks. It was discovered by Barling that the appearance of the ulcers was associated with a pronounced reduction in the total white blood cell count. On one occasion it was 2,600 per cubic millimeter, with 44 per cent polymorphonuclear cells. The ulcers were shallow and appeared on the tongue and inside the mouth and were sufficiently painful to make mastication difficult. There was no response noted to any form of treatment. F. Parks Weber, in discussing this case, refers to 1 previously reported by D. Embleton [202] under the title "Rhythmical Neutropenia with Recurrent Buccal Ulceration" in a woman aged 43. Apparently these cases are similar to those which have been observed in this country and are regarded as instances of a recurrent stomatitis associated with pronounced granulocytopenia. The cause of this condition is not known, but in our minds the possibility that it is due to a filtrable virus should be considered.

Possible Relation of Folic Acid to Agranulocytosis.—It has been suggested by Goldsmith,

196. Oppikofer, E. K.: Ueber Otitis und Rhinitis necroticans agranulocytotica, Schweiz. med. Wchnschr. **73**:107, 1943.

197. Estol Baleztena, M. M.: Agranulocitosis secundaria a un tratamiento de sales de oro, Prensa méd. argent. **31**:85, 1944.

198. Rawls, W. B.: Agranulocytopenia in Patient Receiving Cinchophen and Synthetic Vitamin K, New York State J. Med. **44**:626, 1944.

199. Béguez César, A.: Neutropenia crónica maligna familiar con granulaciones atípicas de les leucocitos, Bol. Soc. cubana de pediat. **15**:900, 1943.

200. Staub, H., and Bucher, K.: Zum Mechanismus der Leukopenia nach i. v. Glykogenzufuhr; Abhängigkeit der Leukopenie von der Grösse der Makromolekel, Schweiz. med. Wchnschr. **73**:904, 1943.

201. Barling, B.: Three and a Half Years' Treatment with Sulphapyridine in Case of Dermatitis Herpetiformis, Lancet **1**:503, 1944.

202. Embleton, D.: Rhythmical Neutropenia with Recurrent Buccal Ulceration, Proc. Roy. Soc. Med. **30**: 980, 1937.

Gordon, Finkelstein and Charipper [203] that since the granulocytopenia produced in rats by sulfonamide compounds can be corrected by liver [204] it appeared logical that liver might have the same effect on the thiourea-induced neutropenia occurring in human beings. The experiments of Goldsmith and his associates were performed as follows: Adult male rats were fed a standard laboratory ration containing 0.5 per cent thiourea for fifty-eight days. Six animals of another group were treated similarly but in addition received 5 per cent of solubilized liver incorporated into the ration. The most striking hematologic change in those animals receiving only thiourea and the stock ration was the pronounced neutrophilic granulocytopenia, which became progressively more severe. It was clearly demonstrated in the second group of rats, however, that the administration of liver almost completely prevents the development of the neutropenia. It is strongly suggested, therefore, that the granulocytopenia induced by the administration of either the sulfonamide compounds or thiourea may be prevented by feeding liver. A norite eluate fraction of liver has been found to alleviate the leukopenia in monkeys maintained on a synthetic diet by Waisman and Elvehjem,[205] and these observers have proposed that folic acid is the active agent in this liver fraction. It appears logical, therefore, to use either folic acid or liver in the prevention and treatment of the granulocytopenia resulting from the administration of thiourea compounds in hyperthyroid patients.

It is stated by Endicott, Kornberg and Daft [206] that in a previous report from the United States Public Health Laboratory the lesions found in rats fed sulfaguanidine in purified diets were described. One of these lesions was agranulocytic aplasia of the bone marrow. During the past year, studies have been made of sulfathiazole, sulfadiazine, sulfanilamide, sulfapyrazine, sulfamerazine and acetylsulfadiazine given rats in purified diets. Among the pathologic changes noted was a depletion of mature granulocytes in the bone marrow with or without an increase in nucleated red blood cells. It is interesting to note that hyperplasia of the marrow was found regularly in rats recovering from granulocytopenia thus induced and that recovery followed treatment with certain liver concentrates.

Totter and his associates [207] have observed that the rhesus monkey requires a water-soluble substance or substances to prevent leukopenia, anemia, diarrhea, lesions of the mouth and colon and eventual death. This factor, for which the term vitamin M was suggested, was shown to be distinct from thiamine, riboflavin and nicotinic acid. Evidence is presented to indicate that the distribution of the factors stimulating growth of Streptococcus lactis R (folic acid) is different from that of factors stimulating growth of vitamin M. Furthermore, they showed that pantothenic acid, choline, paraaminobenzoic acid, pyridoxine and inositol do not prevent nutritional cytopenia in the monkey. They found that the treatment of the cytopenic monkeys with synthetic xanthopterin was followed by a reticulocyte response and an increase in the red and white blood cell count. They concluded that the evidence suggests that xanthopterin or some closely allied substance may be required by the monkey for normal hemocytopoiesis. However, it seems probable that unidentified substances may also be necessary.

It is reported by Wright and Welch [208] that the feeding of a highly purified diet adequate in those members of the vitamin B complex required for the production of excellent growth in rats caused a decided reduction in the hepatic stores of folic acid and biotin compared to the amounts of these factors found in the livers of animals maintained on stock rations. The hepatic storage of these factors was further reduced by the incorporation of succinylsulfathiazole into such synthetic rations. It is important to note that there was no impairment in the storage of riboflavin and of nicotinic acid. These experiments are of interest to us because the findings are in accord with the hypothesis that folic acid deficiency may contribute to the development of agranulocytosis. This is due to the fact that

203. Goldsmith, E. D.; Gordon, A. S.; Finkelstein, G., and Charipper, H. A.: Suggested Therapy for Prevention of Granulocytopenia Induced by Thiourea, J. A. M. A. **125**:847 (July 22) 1944.

204. Spicer, S. S.; Daft, F. S.; Sebrell, W. H., and Ashburn, L. L.: Prevention and Treatment of Agranulocytosis and Leukopenia in Rats Given Sulfanilylguanidine or Succinyl Sulfathiazole in Purified Diets, Pub. Health Rep. **57**:1559, 1942. Kornberg, A.; Daft, F. S., and Sebrell, W. H.: Production and Treatment of Granulocytopenia and Anemia in Rats Fed Sulfonamides in Purified Diets, Science **98**:20, 1943.

205. Waisman, H. A., and Elvehjem, C. A.: Role of Biotin and "Folic Acid" in Nutrition of Rhesus Monkey, J. Nutrition **26**:361, 1943.

206. Endicott, K. M.; Kornberg, A., and Daft, F. S.: Lesions in Rats Given Sulfathiazole, Sulfadiazine, Sulfanilamide, Sulfamerazine, Sulfapyrazine, or Acetylsulfadiazine in Purified Diets, Pub. Health Rep. **59**:49, 1944.

207. Totter, J. R.; Shukers, C. F., and others: Studies on the Relation Between Vitamin M, Xanthopterin and Folic Acid, J. Biol. Chem. **152**:147, 1944.

208. Wright, L. D., and Welch, A. D.: Folic Acid, Biotin and Pantothenic Acid Deficiency and Liver Storage of Various Vitamins in Rats Fed Succinylsulfathiazole in Highly Purified Rations, J. Nutrition **27**:55, 1944.

there may be either a deficiency of folic acid in the diet or perhaps a failure of the intestinal flora to synthesize folic acid, and hence the animal organism is deprived of the factor which controls the maturation of the white blood cells. While this theory is attractive and while there is evidence in favor of it, there are many gaps in the knowledge relating to this process which require further investigation.

It has been pointed out by Waisman and his associates [209] that young monkeys fail to grow and develop on a diet consisting of sucrose, purified casein, mineral salts, corn oil and cod liver oil, together with adequate levels of thiamine, riboflavin, nicotinic acid, pantothenic acid, pyridoxine, choline, paraaminobenzoic acid, and *i*-inositol. When such a diet was given to these animals there was a gradual loss of weight followed by a syndrome of anorexia, leukopenia, lowered resistance to secondary infections, increasing cachexia and death. They demonstrated further that the addition of liver extract, whole liver or "solubilized" liver to the synthetic diet caused a disappearance of all symptoms and that the animals showed striking gains in weight.

Treatment of Agranulocytosis.—An important report is made by Smith, Cohen and Nichols [210] regarding the successful treatment of 2 patients with agranulocytosis with penicillin. They state that up to the time of their publication 1 case in which a similar treatment had been used had been cited.[211] In both the patients classic evidence of agranulocytosis developed after the administration of oxophenarsine hydrochloride (Mapharsen) for the treatment of syphilis. One patient received penicillin in doses of 20,000 Oxford units intramuscularly every three hours, beginning on the third day of the agranulocytosis. Sixteen hours after the first dose the temperature was normal, and the patient made a prompt recovery. A second patient was given penicillin therapy in a dosage of 10,000 units intramuscularly every three hours. Within thirty-eight hours he was afebrile. Although both patients received additional therapy, including blood transfusions, pentnucleotide and yellow bone marrow, it is the opinion of the authors that the striking improvement which occurred during the first twenty-four hours of penicillin therapy warrant its further trial in the treatment of agranulocytosis. To us, the use of penicillin in the treatment of agranulocytosis seems to be logical, because it is the most effective agent available for the control of the sepsis which is the cause of death. Furthermore, penicillin is not responsible for the production of agranulocytosis or of any other untoward results.

The case of a gunner's mate, 24 years old, in whom agranulocytosis developed after the use of sulfathiazole is reported by Meredith, Douglas and Fink.[212] There are two points of interest in the paper. In the first place, the dangers of taking sulfonamide drugs indiscriminately are emphasized. For example, this patient two months before being seen by the authors had been ill for a week with jaundice and did not seek medical attention but took a total of 32 tablets (16 Gm.) of sulfathiazole during the course of the illness. Four years before he had been uneventfully treated with sulfathiazole for gonorrhea and since that time had frequently taken smaller amounts for minor infections and venereal prophylaxis. The present illness began when the patient experienced the symptoms of a cold and on his own initiative took 12 tablets (6 Gm.) of sulfathiazole during the day. The following day his throat was definitely sore, and that evening he had chills followed by fever. When seen at that time his white blood cell count was 2,700 per cubic millimeter, with a differential count of 56 per cent polymorphonuclear leukocytes, 40 per cent lymphocytes, 3 per cent monocytes and 1 per cent eosinophils. The following day the white blood cell count was 1,500 per cubic millimeter, with 100 per cent lymphocytes. The second point of interest is the use of large doses of penicillin as one form of treatment. When the diagnosis of agranulocytosis was established, intensive therapy was immediately instituted, consisting of transfusions of whole blood, intramuscular injections of liver extract, injections of pentnucleotide and the use of penicillin. The latter was administered in quantities of 100,000 units for the initial dose, 120,000 units during each of the following six days and then 90,000, 40,000 and 20,000 units on subsequent days. Penicillin therapy was discontinued on the eleventh day. In résumé, the therapy over a period of ten days was as follows: 2,500 cc. of whole blood, 260 units of liver extract, 270 cc.

209. Waisman, H. A.; Rasmussen, A. F., Jr.; Elvehjem, C. A., and Clark, P. F.: Studies on Nutritional Requirements of Rhesus Monkey, J. Nutrition **26**:205, 1943.

210. Smith, L. B.; Cohen, F., and Nichols, R. G.: Agranulocytosis Treated with Penicillin, J. A. M. A. **126**:1027 (Dec. 16) 1944.

211. Keefer, C. S., and others: Penicillin in the Treatment of Infections: Report of Five Hundred Cases; Statement by the Committee on Chemotherapeutic and Other Agents, Division of Medical Sciences, National Research Council, J. A. M. A. **122**:1217 (Aug. 28) 1943.

212. Meredith, W. C.; Douglas, A. H. R., and Fink, H.: Penicillin in Malignant Granulocytopenia: Report of Case, U. S. Nav. M. Bull. **43**:1017, 1944.

of pentnucleotide and 970,000 Oxford units of penicillin. The patient made a complete recovery.

The case of a 22 year old seaman in whom agranulocytosis developed and who recovered after the use of penicillin is reported by Sprague and Ferguson.[213] This patient six months before had been treated for six weeks with a sulfonamide drug for gonorrhea. When observed by the authors, the patient again had received 36 Gm. of sulfathiazole over a period of six days. At the end of this time fever developed; there was ulceration of the throat, and the white blood cell count was found to be 900 per cubic millimeter. It later fell to 275. The patient received many forms of treatment, including twenty-five blood transfusions of 250 cc. each, pentnucleotide, yellow bone marrow and liver extract. In addition, penicillin was given in amounts of 20,000 units every four hours intramuscularly for five days. After this its use was discontinued and later resumed. A total dose of approximately 6,480,000 units was given, to which the authors attribute this person's recovery.

Pyridoxine was introduced in the treatment of leukopenia and granulocytopenia, according to Cantor and Scott,[214] for the following reasons: First, it had been shown that folic acid, a component of the vitamin B complex, is effective in preventing and curing the neutropenia produced in rats by the feeding of the insoluble sulfonamide compounds. Additional work has indicated that this effect is probably an indirect one, in the sense that the folic acid fed is required by coliform bacteria in the intestines for the production of some accessory substance, which, in turn, produces a granulocytic response. It has also been observed that microcytic hypochromic anemia produced in dogs by a deficiency of vitamin B_6 is not relieved by the administration of iron. Furthermore, the anemia of pellagra and pernicious anemia are similarly unresponsive to iron, while pyridoxine is a constitutent of liver and yeast, both of which are effective in the treatment of these disorders. Also, it has been reported that the administration of pyridoxine intravenously to pellagrins and to patients with pernicious anemia in a state of relapse results in improvement within forty-eight hours, with a striking increase in the leukocyte count, especially in the granulocytic series. Furthermore, they state that the administration of pyridoxine produces some benefit in persons with Cooley's anemia when it is used in association with chorionic gonadotropine. While all these observations may be true, it is not clear to us why pyridoxine should be of value in the treatment of leukopenia and granulocytopenia. The observers treated 3 patients with agranulocytic angina with a 10 per cent solution of pyridoxine hydrochloride in isotonic solution of sodium chloride in amounts totaling 125 mg. to 200 mg. intravenously daily. In 1 of these patients agranulocytic angina had developed after the administration of 4 Gm. of sulfathiazole given over a period of twenty-four hours. There was no medication given to the second patient which could have been responsible for the agranulocytic angina except self-administered acetylsalicylic acid. The condition developed in the third patient after the use of thiouracil about two months after this therapy for hyperthyroidism was instituted. A total of 13.5 Gm. of the drug had been taken. It is unusual, to say the least, that agranulocytic angina should develop in a patient who had received 4 Gm. of sulfathiazole and also that the same condition should develop in a patient who had received only a total of 13.5 Gm. of thiouracil over a period of two months. It is stated that the temperature of each patient fell to normal limits and that the symptoms disappeared promptly after the use of pyridoxine hydrochloride. These occurences were associated with an increase in the number of leukocytes and the reappearance of granulocytes in the blood. Recovery followed in all patients. The observers conclude by stating that pyridoxine hydrochloride administered intravenously appears to be a useful agent for the treatment of agranulocytic angina of toxic origin. Its effectiveness in 3 instances suggests that pyridoxine acts by direct stimulation of the myelocytic elements in the bone marrow. No one can deny the possibility that pyridoxine hydrochloride may be effective in treating this disease. On the other hand, the recovery of 3 patients from a disease in which spontaneous recovery is not rare after the use of a drug for whose use there is no special logical basis does not place the claim on secure grounds.

213. Sprague, H. B., and Ferguson, L. K.: Agranulocytosis Treated with Penicillin: Report of Case, U. S. Nav. M. Bull. **43**:1014, 1944.

214. Cantor, M. M., and Scott, J. W.: Effect of Vitamin B_6 (Pyridoxine) in Treatment of Leukopenia and Granulocytopenia of Toxic Origin in Humans: Preliminary Report, Science **100**:545, 1944.

(To Be Continued)

Book Reviews

Constitution and Disease. By Julius Bauer, M.D. Price, $4. Pp. 247. New York: Grune & Stratton, Inc., 1945.

In this book Dr. Bauer tries, one might say, to bridge the gap between pure genetics and clinical medicine. The general question of constitution is discussed from various aspects, with liberal allusion to practical examples. Obviously a discussion of this sort must be written from a somewhat personal point of view, and not every one will agree in all details with Dr. Bauer's conclusions. From any standpoint, however, the book furnishes interesting and stimulating reading.

Clinical Roentgenology of the Digestive Tract. By Maurice Feldman, M.D., assistant professor of gastroenterology, University of Maryland. Second edition. Price, $7. Pp. 769, with 551 illustrations. Baltimore: Williams & Wilkins Company, 1945.

In THE ARCHIVES (**53**:1237 [June] 1939) was reviewed the first edition of this book, the reviewer commending the author on his complete and understanding presentation of such an extensive subject. The second edition is even more praiseworthy and, indeed, sets an unusual example in that it is shorter, cheaper and more extensively illustrated than was its predecessor. Most second editions do not follow such a pattern.

The text continues to present a thorough consideration of gastrointestinal disease which will interest both radiologist and clinician. The illustrations are admirable. The pertinent literature is extensively reviewed, and there is a short up-to-date bibliography at the end of each chapter. The present reviewer is glad to reaffirm in THE ARCHIVES that the book is an invaluable reference work and to predict for it continued success.

Penicillin Therapy, Including Tyrothricin and Other Antibiotic Therapy. By John A. Kolmer, M.S., M.D., Dr.P.H., Sc.D., LL.D., L.H.D., F.A.C.P.; professor of medicine in the School of Medicine and the School of Dentistry, Temple University; Director of the Research Institute of Cutaneous Medicine; formerly professor of pathology and bacteriology, Graduate School of Medicine, University of Pennsylvania. Cloth. Price not given. Pp. 302, with 42 illustrations and tables. New York: D. Appleton-Century Company, Inc., 1945.

This is a timely book and will interest practitioners of medicine from all branches. It represents the author's personal experience in the use of penicillin, together with a reasonably complete review of the work of other clinicians and investigators. It deals with the structure of penicillin and the method of its production and assay. It also describes the method of determining the concentration of penicillin in the blood, a procedure which is not generally carried out. The various methods of administration of penicillin are adequately dealt with, and a special section is included to cover the use of penicillin and its related substances in dentistry and oral surgery. In addition to penicillin, tyrothricin and gramicidin and some other substances that have come to be known as antibiotics are discussed. An appendix is included to care for matter that has come to the author's attention since the original manuscript was completed.

The book is a most interesting and valuable one. As with all books on new therapeutic agents, some of the statements in this book have already been modified by experience, but there is no detraction from the general value of the book on that account.

The bibliography and the list of references are excellent.

The Specificity of Serological Reactions. By Karl Landsteiner, M.D. Second revised edition. Price, $5. Pp. 310. Cambridge, Mass.: Harvard University Press, 1945.

In 1936 the late Dr. Landsteiner published the first edition of his now classic monographic review of serologic reactions. The advances of the subsequent years made a revision desirable, and the text was completed at the time of Dr. Landsteiner's death; we are indebted to his son Ernest, also a physician, who undertook to see the revision carried through to the state of final publication.

This volume contains an excellent chapter on molecular structure and intermolecular forces as they apply to immunologic reactions, contributed by Dr. Linus Pauling. The remainder of the book includes an introductory section defining immunologic concepts and terminology and chapters on the serologic properties of various types of proteins, nonprotein cell substances and artificial conjugated antigens derived from simple chemical compounds, on cell antigens, on the nature and specificity of antibodies and on antigen-antibody reactions.

The enormous body of pertinent literature has been analyzed and built into a coherent concept of immunologic specificity by the worker whose wide-ranging intellect and personal contributions to fundamental research in the field made him uniquely fitted for the task. All who are interested in immunologic reactions are indebted to Dr. Karl Landsteiner for this summary of his interpretation of the existing knowledge on the subject and will treasure the book as they do the memory of his scientific achievements.

Publicaciones del centro investigaciones tisiológicas. Edited by Prof. Roque A. Izzo, Director. Volume 7. Buenos Aires, Argentina: Pabellon "Las Provincias," Hospital Tornu, 1944.

Like the preceding six volumes, this (the seventh) volume of the collected studies of the Centro de Investigaciones tisiológicas consists of a group of papers and monographs on thoracic conditions. Although some of the work is of a high order, it would be expecting too much to have every study a classic. The monograph on constitution and tuberculosis is one of the best reviews on the subject and is supported by a worthy original study. There is little doubt left in the reader's mind that some genotypic (hereditary) aspects of constitution determine whether certain patients, such as the "heavy-set" and "elongated" types, present different reactions to the tubercle bacillus, with different prognoses. The former is found to have a

good prognosis and the latter a bad one, a distinction conforming to wide clinical experience. There are also different reactions in different races and ages of people to tuberculous infection, although the explanation offered for these reactions are sometimes inadequate and the authorities cited at no time present a majority opinion on any phase of the subject. The attempt to assign various "endocrine types of habitus" to special types of tuberculosis results in spreading the evidence rather thin. There is not due consideration of the possibilities of counterbalance of one or more of the many unfavorable characteristics by other more favorable characteristics.

A monograph on malformations of the lungs and bronchi is also complete and well presented, but the work has the tendency of other similar studies to fail to distinguish with certainty the acquired disease from true congenital cystic disease of the lung. Another article points out the necessity of performing tuberculin tests and examination of sputum in addition to roentgenographic examinations of the chest for the diagnosis of tuberculosis in children. Another, beautifully illustrated, article is devoted to the cervicothoracic diaphragm, or endothoracic fascia, covering the dome of the lungs. Still another study worthy of mention is one of pulmonary alveolar cells.

The paper and print are of the best. The illustrations are good in some articles and poor in others.

The volume is highly recommended for persons interested in pulmonary diseases.

Diseases of the Nervous System in Infancy, Childhood and Adolescence. By Frank R. Ford, M.D. Second edition. Cloth. Price, $12.50. Pp. 1,143, with illustrations. Springfield, Ill.: Charles C Thomas, Publisher, 1944.

Dr. Ford allowed eight years to elapse before bringing out his present (second) edition, and it would require much more space to review in detail such an encyclopedic treatise. Features to be specially noted are the introductory chapters on the examination of the nervous system in general and the clinical aspects of the anatomy and physiology of the nervous system, the many excellent photographs and diagrams, the thorough bibliographies and the excellent general format with large readable type. Every variation of the complicated neurologic disorders of childhood is mentioned, as, for example, no less than forty-three heredofamilial and degenerative diseases. Prenatal diseases, infections, tumors and so forth present a hardly less formidable array. The illustrative case histories seem especially useful in dealing with material of this sort in which the nosologic aspect is so important. Dr. Ford's treatise will undoubtedly remain the standard reference work on the subject.

Military Medical Manuals—A Manual of Tropical Medicine. Prepared under the auspices of the Division of Medical Sciences of the National Research Council. Colonel Thomas T. Mackie, M.C., A.U.S.; Major George W. Hunter III, Sn.C., A.U.S., and Captain C. Brooke Worth, M.C., A.U.S. Price, $6. Pp. 727, with 284 illustrations. Philadelphia and London: W. B. Saunders Company, 1945.

Tropical medicine is a singular specialty because it is a conglomerate of sections from nearly all the medical and biologic sciences. Tropical medicine deals with the medical problems of the earth's equator. The physician in the tropics, therefore, must know something of nearly everything; he must be protozoologist, bacteriologist, clinician, laboratory technician, hygienist, entomologist, chemist, expert in virus, rickettsial and metabolic diseases, helminthologist, drainage engineer, rat exterminator, administrator and surgeon. To provide him with a portable text dealing with these subjects in acceptable detail would be impossible. A manual, therefore, must limit itself to the most important topics likely to be required by the physicians actually in the tropics.

How well this manual has achieved its design to provide a concise and dependable source of old and new information is best comprehended by a careful study of the book. The numerous diseases and hazards of the tropics are presented briefly and clearly. The recommended treatments include the recent advances in medicine. Numerous instructive sketches and unusually clear illustrations illuminate the text. Many tables which condense parallel data are included. There are sections devoted to medically important arthropods and to medically important animals. Nutritional diseases and the effects of inadequate diets are discussed. Diagnostic laboratory procedures are carefully outlined. Finally, there is a generous index.

The manual kindled the enthusiasm of this reviewer. It is not only the best book of its kind; it is a combination of the good features of all the books of its kind. Here in one source the physician in the tropics can find practically everything he may need, presented in clear and authoritative fashion. The authors made use of all the sources available, as the long list of acknowledgments will prove; but the excellent condensation of this information is an achievement worthy of lavish praise. It is especially interesting to read of tsutsugamushi disease (scrub typhus) in New Guinea, of the uses of dimethylphthalate and of DDT in combating insects and of the usefulness of sulfonamide compounds in plague and dysenteries.

It is difficult to find something to criticize in this manual. One may wish that more had been said of the uses of penicillin in the treatment of certain infections, such as that caused by Streptobacillus moniliformis, for example. It would be valuable to present in a manual to be used in all countries tables of weights and measures, with instructions on how to convert Centigrade readings to Fahrenheit, inches to centimeters and liters to pints. Also, a brief glossary of terms, especially of the names given by the British to the common useful drugs, would be very helpful. But these are small matters. There are no typographic errors to be found, and the technical work of printing and binding is excellent. This manual is highly recommended to all who have an interest in tropical medicine. It will be found especially useful to medical students.

News and Comment

GENERAL NEWS

The American College of Physicians. — The American College of Physicians will hold its annual meeting May 13-17, 1946 at the Philadelphia Municipal Auditorium, Thirty-Fourth Street below Spruce Street. Further information may be procured from the president, Dr. Ernest E. Irons, Chicago, or from the general chairman, Dr. George Morris Pierson, Philadelphia.

INDEX TO VOLUME 76

Book Reviews are grouped together and are indexed under that heading in alphabetical order under the letter B.

Abnormalities and Deformities: See under names of organs and regions
Abscess: See under names of organs and regions, as Liver; Lungs; etc.
Acacia: See also Nephritis
effect of intravenously administered solution of acacia on animals, 39
Achlorhydria: See Stomach
Acid, Ascorbic: See Ascorbic Acid
Cevitamic: See Ascorbic Acid
Folic: See Vitamins, B
Nicotinic: See Nicotinic Acid
Acrosclerosis: See Scleroderma
Actinomyces; streptomycin and other antibodies, 119
streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Adrenals; small adenomas of adrenal cortex in hypertension and diabetes, 284
Aeronautics: See Aviation and Aviators
Agranulocytosis due to thiouracil, 387
possible relation of folic acid to, 393
toxic effect of sulfonamide drugs including agranulocytosis, 380
treatment of, 395
Air, control of air-borne infections, 122
Air Passages: See Respiratory Tract
Amebiasis; amebic pericarditis, 88
American College of Physicians, 398
Anemia, Agranulocytic: See Agranulocytosis
and nutrition, 360
aplastic, 379
associated with pregnancy, 362
erythroblastic; Mediterranean anemia, 375
experimental, and iron metabolism, 358
hemolytic, 363
macrocytic; pernicious anemia and related macrocytic anemias, 60
macrocytic, related to pernicious anemia, 247
Mediterranean: See Anemia, erythroblastic
pernicious, 239
pernicious, achlorhydria in, 242
pernicious, and related macrocytic anemias, 60
pernicious, diagnosis of, 245
pernicious, in Chinese, 130
pernicious, nervous system in cases of, 245
pernicious, related to macrocytic anemia, 247
sensitivity to liver extract, 252
sickle cell; habitus of patients with active sickle cell anemia of long duration, 47
sickle cell, in white patients with ulcers of ankles; report of 2 cases, 230
treatment, 248
Angina, Agranulocytic: See Agranulocytosis
Vincent's: See Fusospirochetosis
Angiotonin; studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
Ankles, Ulcers: See Ulcers
Antibiotics; streptomycin and other antibiotics, 119
Antibodies: See Antigens and Antibodies
Antigens and Antibodies; antibody formation, 187
Armed Forces Personnel: See Aviation and Aviators; Military Medicine; etc.
Armies: See Military Medicine
Arteries: See Arteriosclerosis; Blood pressure; Embolism; Extremities, blood supply; Thrombosis; Vasomotor System; etc.
Coronary: See Coronary Vessels
Arteriosclerosis; arteriosclerotic peripheral vascular disease in diabetes, 34
Arthritis, rheumatoid, 183
rheumatoid, characteristic vascular pattern in patients with, 139
Ascorbic Acid: congenital idiopathic methemoglobinemia; favorable response to ascorbic acid therapy, 299
Atherosclerosis: See Arteriosclerosis
Atmosphere: See Air
Atrophy: See under Tongue
Aviation and Aviators; paroxysmal hemoglobinuria due to cold hemolysin; observations with report of case of its occurrence in aerial gunner, 292

Bacilli: See Bacteria
Bacteria: See also Meningococci; Streptococci; Viruses; etc.
Abortus and Melitense Group: See Undulant Fever
bacteriologic studies of resistance to penicillin, 118
Diphtheria: See Diphtheria
Dysentery: See Dysentery
Eberth's: See Typhoid
Influenza: See Influenza
Leprosy: See Leprosy
Shigella: See Dysentery
Streptococci: See Streptococci
Tularense: See Tularemia
Baker, A. B.: Changes in central nervous system associated with encephalitis complicating pneumonia; clinical study, 146
Bartlett, C. J.: Extragenital chorionepithelioma in male, 347
Benzene; toxic effects of industrial poisons, 376
Berg, M.: Acute porphyria; clinical and pathologic observations, 335
Beriberi: relation of nutritional deficiency to cardiac dysfunction, 341
Bethell, F. H.: Blood; review of recent literature, 239, 358
Bierring, W. L.: Sickle cell anemia in white patients with ulcers of ankles; report of 2 cases, 230
Bilharziasis: See Schistosomiasis
Biliary Tract: See Gallbladder; Liver
Binger, M. W.: Effect of intravenously administered solution of acacia on animals, 39
Blood: See also Erythrocytes; Hemoglobin and Hemoglobin Compounds
Circulation: See also Heart; Vasomotor System; etc.
circulation in acute myocardial infarction, 54
coagulation; gallbladder dye (iodophthalein sodium); effect of intravenous injections on coronary flow, blood pressure and blood coagulation, 143
Diseases: See Anemia; etc.
plasma quinacrine concentration in treatment of plasmodium vivax malaria acquired in South Pacific, 352
pressure; gallbladder dye (iodophthalein sodium); effect of intravenous injections on coronary flow, blood pressure and blood coagulation, 143
pressure, high; small adenomas of adrenal cortex in hypertension and diabetes, 284
pressure, high; studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
review of recent literature, 239, 358
Blumberg, R. W.: Epidemic hepatitis with and without jaundice; some clinical studies on 255 patients among troops in combat zone, 102; correction, 188
Blumenthal, H. T.: Small adenomas of adrenal cortex in hypertension and diabetes, 284
Body, habitus of patients with active sickle cell anemia of long duration, 47
Boisvert, P. J.: Etiology and pathogenesis of rheumatic fever, 131
Hemolytic streptococcus sore throat; detailed study of simultaneous infection of large number of men by single type, 278

Bollman, J. L.: Effect of intravenously administered solution of acacia on animals, 39
Bones: See under names of bones

Book Reviews:
Arterial Hypertension: Its Diagnosis and Treatment; I. H. Page and A. C. Corcoran, 255
Biological Symposia, Volume XI, Aging and Degenerative Diseases; edited by R. A. Moore, 256
Clinical Case-Taking; G. R. Herrmann, 130
Clinical Heart Disease; S. A. Levine, 177
Clinical Roentgenology of Digestive Tract; M. Feldman, 397
Constitution and Disease; J. Bauer, 397
Diseases of Nervous System in Infancy, Childhood and Adolescence; F. R. Ford, 398
Doctors at War; edited by M. Fishbein, 177
Doctor's Job; C. Binger, 62
Duodenal and Jejunal Peptic Ulcer; R. Nissen, 130
Essentials of Allergy; L. H. Criep, 130
Examination of Reflexes; R. Wartenberg, 256
Familial Susceptibility to Tuberculosis: Its Importance as Public Health Problem; R. R. Puffer, 62
Hipertensión arterial nefrógena; E. Braun-Menéndez and others, 62
Hypertension and Hypertensive Disease; W. Goldring and H. Chasis, 61
Microbial Antagonisms and Antibiotic Substances; S. A. Waksman, 255
Military Medical Manuals—Manual of Clinical Mycology; Division of Medical Sciences of National Research Council, 256
Military Medical Manuals—Manual of Tropical Medicine: Prepared under auspices of Division of Medical Sciences of National Research Council; T. T. Mackie, G. W. Hunter III and C. B. Worth, 398
Patients Have Families; H. B. Richardson, 177
Penicillin and Other Antibiotic Agents; W. E. Herrell, 255
Penicillin Therapy, Including Tyrothricin and Other Antibiotic Therapy; J. A. Kolmer, 397
Poet Physicians: Anthology of Medical Poetry Written by Physicians; compiled by M. L. McDonough, 256
Publicaciones del centro investigaciones tisiológicas; edited by R. A. Izzo, 397
Specificity of Serological Reactions; K. Landsteiner, 397
Trauma in Internal Diseases; R. A. Stern, 256
Trichinosis; S. E. Gould, 61

Bornstein, J.: Clinical syndrome in patients with pulmonary embolism, 234
Brain: See also Meninges; Nervous System; etc.
Diseases: See Encephalitis
neurologic complications during meningococcic meningitis treated with sulfonamide drugs, 201
Bright's Disease: See Nephritis
Bronchitis; ulcerative tracheobronchitis following atypical pneumonia; report of cases, 93
Brucellosis: See Undulant Fever
Burch, G. E.: Habitus of patients with active sickle cell anemia of long duration, 47
Rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
Study of water and heat loss from respiratory tract of man; methods: gravimetric method for measurement of rate of water loss; quantitative method for measurement of rate of heat loss, 308

Cancer: See under names of organs and regions
Cardiovascular System: See also Heart; Vasomotor System; etc.
relation of nutritional deficiency to cardiac dysfunction, 341
Cateno, C. F.: Plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
Cerebrospinal Fever: See Meningitis
Cerebrum: See Brain
Chagas' Disease: See Trypanosomiasis
Chemotherapy, 120. See also under names of diseases
Chernoff, H. M.: Extragenital chorionepithelioma in male, 347
China and Chinese; pernicious anemia in Chinese, 130
Cholera, 180
Chorionepithelioma, extragenital, in male, 347
Circulation: See Blood, circulation
Circulatory System: See Blood, circulation; Heart
Climate; rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
Coccidioidomycosis, 128
Cold; paroxysmal hemoglobinuria due to cold hemolysin; observations with report of case of its occurrence in aerial gunner, 292
Cold Hemagglutination: See Pneumonia
Colds: See under Respiratory Tract
Colitis, ulcerative, 180
Collen, M. F.: Optimum dose of sulfadiazine in treatment of pneumococcic pneumonia, 22
Communicable Diseases: See also Measles; Meningitis; Syphilis; etc.
infectious diseases; eleventh annual review of significant publications, 114, 178
Comroe, B. I.: Characteristic vascular pattern in patients with rheumatoid arthritis, 139
Cooley's Anemia: See Anemia, erythroblastic
Coronary Vessels; gallbladder dye (iodophthalein sodium); effect of intravenous injections on coronary flow, blood pressure and blood coagulation, 143
Cutting, W. C.: Sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161
Cysts: See under names of organs and regions

Darling's Disease: See Histoplasmosis
Death, account of stock, 210
sudden; clinical syndrome in patients with pulmonary embolism, 234
Deficiency Diseases: See also Nutrition; Vitamins; etc.
relation of nutritional deficiency to cardiac dysfunction, 341
Dengue, 186
Dentition: See Teeth
Diabetes, Bronze: See Hemochromatosis
Diabetes Mellitus; arteriosclerotic peripheral vascular disease in diabetes, 34
small adenomas of adrenal cortex in hypertension and diabetes, 284
Diarrhea: See also Dysentery
epidemic nausea, vomiting and diarrhea (viral or viroid dysentery), 178
Dick, W. S.: Sickle cell anemia in white patients with ulcers of ankles; report of 2 cases, 230
Diet and Dietetics: See Nutrition; Vitamins; etc.
Diphtheria, 128
Dye: See Gallbladder
Dysentery: See also Diarrhea
Amebic: See Amebiasis
epidemic nausea, vomiting and diarrhea (viral or viroid dysentery?), 178
shigellosis, 179

Edema: See also under names of organs
Nephrotic: See Nephritis
powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
Electrocardiogram: See under Heart
Elias, W. F.: Streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Ellerbrook, L. D.: Plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
Elliptocytosis: See Erythrocytes
Embolism, pulmonary, clinical syndrome in patients with, 234
Encephalitis, 182
changes in central nervous system associated with encephalitis complicating pneumonia; clinical study, 146
Endocarditis, subacute bacterial, healed, 1
subacute bacterial, penicillin for, 115
Energy Exchange: See Metabolism
Erythroblastosis: See Anemia, erythroblastic

Erythrocytes: See also Anemia; Hemoglobin and Hemoglobin Compounds; Hemolysis; etc.
ellipsocytosis, 373
Esophagus; esophageal lesions associated with acrosclerosis and scleroderma, 189
Evans, T. S.: Extragenital chorionepithelioma in male, 347
Ewing, P. L.: Studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
Extremities, blood supply; arteriosclerotic peripheral vascular disease in diabetes, 34
blood supply; characteristic vascular pattern in patients with rheumatoid arthritis, 139
Paralysis: See Poliomyelitis

Farmer, T. W.: Neurologic complications during meningococcic meningitis treated with sulfonamide drugs, 201
Favre-Nicolas Disease: See Lymphogranuloma Venereum
Feces, fat in; powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Fever: See also Malaria; Typhoid; Typhus; Undulant Fever; etc.
clinical syndrome in patients with pulmonary embolism, 234
Desert: See Coccidioidomycosis
Malta: See Undulant Fever
Rocky Mountain Spotted: See Rocky Mountain Spotted Fever
Undulant: See Undulant Fever
Valley: See Coccidioidomycosis
Filariasis, 186
Finks, R. M.: Epidemic hepatitis with and without jaundice; some clinical studies on 255 patients among troops in combat zone, 102; correction, 188
Fitz, R.: Account of stock, 210
Food: See Nutrition; Vitamins; etc.
Freezing: See Cold
Frei Reaction: See Lymphogranuloma Venereum
Fusospirochetosis; Vincent's infection, 180

Gallbladder dye (iodophthalein sodium); effect of intravenous injections on coronary flow, blood pressure and blood coagulation, 143
Gillman, J.: Powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
Gillman, T.: Powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
Gingivostomatitis: See Fusospirochetosis
Glomerulonephritis: See under Nephritis
Gordon, H. H.: Plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
Granuloma, Coccidioidal: See Cococcidioidomycosis inguinale, 187
Tropicum: See Frambesia
Venereal: See Lymphogranuloma Venereum
Gray, S. H.: Small adenomas of adrenal cortex in hypertension and diabetes, 284
Gregory, R.: Studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
Grip: See Influenza
Grossman, J.: Some observations on primary atypical pneumonia, 328

Haemophilus Influenzae: See Influenza
Heart: See also Blood, circulation
absence of electrocardiographic changes in tsutsugamushi fever (scrub typhus); report of 200 consecutive cases, 217
account of stock, 210
circulation in acute myocardial infarction, 54
Diseases: See also Endocarditis; Myocarditis; Pericarditis; etc.
diseases; clinical syndrome in patients with pulmonary embolism, 234
relation of nutritional deficiency to cardiac dysfunction, 341
Heat, rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
study of water and heat loss from respiratory tract of man; methods: gravimetric method for measurement of rate of water loss; quantitative method for measurement of rate of heat loss, 308
Heine-Medin Disease: See Poliomyelitis
Hemagglutination: See Pneumonia
Hematology: See Blood
Hemoglobin and Hemoglobin Compounds: See also Anemia; Blood; Erythrocytes; Hemoglobinuria; Hemolysis
congenital idiopathic methemoglobinemia; favorable response to ascorbic acid therapy, 299
Hemoglobinuria, paroxysmal, due to cold hemolysin; observations with report of case of its occurrence in aerial gunner, 292
Hemolysis: See also Anemia, pernicious
paroxysmal hemoglobinuria due to cold hemolysin; observations with report of case of its occurrence in aerial gunner, 292
Hemostasis: See Blood, coagulation; etc.
Hepatitis: See Jaundice; Liver
Herzstein, J.: Arteriosclerotic peripheral vascular disease in diabetes, 34
Histoplasmosis, 128
Howell, W. L.: Absence of electrocardiographic changes in tsutsugamushi fever (scrub typhus); report of 200 consecutive cases, 217
Hyperpyrexia: See Fever
Hypertensin: See Angiotonin
Hypertension: See Blood pressure, high
Hyperthermia: See under Fever
Hypothermia: See Cold

Icterus: See Jaundice
Industry, toxic effects of industrial poisons, 376
Infantile Paralysis: See Poliomyelitis
Infants, powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
Infarction: See under Heart
Infection: See also under names of bacteria, as Meningococci; Pneumococci; Streptococci; etc.
cold hemagglutination in primary atypical pneumonia and other common infections, 75
streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161
Infectious Diseases: See Communicable Diseases
Influenza, 126
Injections, effect of intravenously administered solution of acacia on animals, 39
intravenous, of acacia; clinical and physiologic effects on patients with nephrotic edema, 167
Insulin: See Diabetes Mellitus
Intestines, Diseases: See Diarrhea; Dysentery
Iodophthalein: See Gallbladder
Iron metabolism and experimental anemia, 358

Japanese River Fever: See Tsutsugamushi Disease
Jaundice, epidemic hepatitis with and without jaundice; some clinical studies on 255 patients among troops in combat zone, 102; correction, 188
epidemic; infectious hepatitis, 178
Jenkins, D. W.: Sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161

Johnson, J. B.: Intravenous injection of acacia; clinical and physiologic effects on patients with nephrotic edema, 167
Jones, J. P.: Cold hemagglutination in primary atypical pneumonia and other common infections, 75

Kay, E. B.: Ulcerative tracheobronchitis following atypical pneumonia; report of cases, 93
Kern, F., Jr.: Amebic pericarditis, 88
Kidneys: See also Urinary Tract
Diseases: See Nephritis
extragenital chorionepithelioma in male, 347
Kirklin, B. R.: Esophageal lesions associated with acroscleresis and scleroderma, 189
Korenberg, M.: Pernicious anemia and related macrocytic anemias, 60
Kyhos, E. D.: Reversal of lingual atrophic changes with nicotinamide therapy, 31

Lawson, F. E.: Gallbladder dye (iodophthalein sodium); effect of intravenous injections on coronary flow, blood pressure and blood coagulation, 143
Leach, J. E.: Pneumothorax in young adult males; descriptive statistics in 126 cases, 264
Leprosy, 187
Levin, W. C.: Studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
Lingua: See Tongue
Lippincott, S. W.: Plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
Liver, amebic abscess; amebic pericarditis, 88
Diseases: See also Jaundice
diseases; epidemic hepatitis with and without jaundice; some clinical studies on 255 patients among troops in combat zone, 102; correction, 188
effect of intravenously administered solution of acacia on animals, 39
powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
sensitivity to liver extract, 252
Loewe, L.: Healed subacute bacterial endocarditis, 1
Lungs: See also Respiratory Tract; etc.
abscess; putrid pulmonary abscess without foul sputum (shut-off pulmonary abscess), 154
Embolism: See Embolism
rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
study of water and heat loss from respiratory tract of man; methods: gravimetric method for measurement of rate of water loss; quantitative method for measurement of rate of heat loss, 308
Tuberculosis: See Tuberculosis, pulmonary
Lymphogranuloma Venereum, recurrent meningoencephalitis due to virus of, 174

Maire, E. D.: Paroxysmal hemoglobinuria due to cold hemolysin; observations with report of case of its occurrence in aerial gunner, 292
Mal del Pinto: See Pinta
Malaria, 186
plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
pneumonia in association with, 126
Malnutrition: See Nutrition
Malta Fever: See Undulant Fever
Marble, A.: Plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
Measles, 184
Medicine, Aviation: See Aviation and Aviators
Military: See Military Medicine
Medin-Heine Disease: See Poliomyelitis
Mediterranean Anemia: See Anemia, erythroblastic
Meninges; tuberculous meningitis, 182
Meningitis: See also Meningococci; Meningoencephalitis
neurologic complications during meningococcic meningitis treated with sulfonamide drugs, 201
streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Tuberculous: See Meninges
tularemic; review of literature and report of case with postmortem observations, 163
Meningococci: See also Meningitis
infections, sulfonamide prophylaxis for, 122
Meningoencephalitis: See also Encephalitis; Meningitis
recurrent, due to virus of lymphogranuloma venereum, 174
Metabolism, rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
study of water and heat loss from respiratory tract of man; methods: gravimetric method for measurement of rate of water loss; quantitative method for measurement of rate of heat loss, 308
Methemoglobinemia: See Hemoglobin and Hemoglobin Compounds
Meyers, M. C.: Blood; review of recent literature, 239, 358
Microfilaria: See Filariasis
Military Medicine: See also Aviation and Aviators; etc.
absence of electrocardiographic changes in tsutsugamushi fever (scrub typhus); report of 200 consecutive cases, 217
epidemic hepatitis with and without jaundice; some clinical studies on 255 patients among troops in combat zone, 102; correction, 188
exotic diseases, 186
plasma quinacrine concentration in treatment of Plasmodium vivax malaria acquired in South Pacific, 352
pneumothorax in young adult males; descriptive statistics in 126 cases, 264
some observations on primary atypical pneumonia, 328
South American trypanosomiasis (Chagas' disease), 219
Miller, H.: South American trypanosomiasis (Chagas' disease), 219
Mites: See Tsutsugamushi Disease
Morphea: See Scleroderma
Moseley, V.: South American trypanosomiasis (Chagas' disease), 219
Mouth, sulfonamide prophylaxis in oral surgery, 121
Mumps: See Parotitis
Myocarditis, acute, in mumps (epidemic parotitis), 257
Myocardium: See Heart; Myocarditis

Naide, M.: Characteristic vascular pattern in patients with rheumatoid arthritis, 139
Nausea; epidemic nausea, vomiting and diarrhea (viral or viroid dysentery?), 178
Nephritis: See also Kidneys, diseases
intravenous injection of acacia; clinical and physiologic effects on patients with nephrotic edema, 167
Nephrosis: See Kidneys, diseases; Nephritis
Nerves: See Nervous System; Paralysis
Nervous System: See also Brain; etc.
changes in central nervous system associated with encephalitis complicating pneumonia; clinical study, 146
diseases of central nervous system, 180
in cases of pernicious anemia, 245
Neuhof, H.: Putrid pulmonary abscess without foul sputum (shut-off pulmonary abscess), 154
Neutropenia: See Agranulocytosis
Newman, L. H.: Intravenous injection of acacia; clinical and physiologic effects on patients with nephrotic edema, 167
Nicolas-Favre Disease: See Lymphogranuloma Venereum
Nicotinic Acid: See also Vitamins, B
reversal of lingual atrophic changes with nicotinamide therapy, 31
Nitrotoluene; trinitrotoluene poisoning, 378

Noran, H. H.: Changes in central nervous system associated with encephalitis complicating pneumonia; clinical study, 146
Nutrition: See also Diet and Dietetics; Vitamins; etc.
and anemia, 360

Occupations: See Industry
O'Leary, P. A.: Esophageal lesions associated with acroscleroses and scleroderma, 189
Olsen, A. M.: Esophageal lesions associated with acroscleroses and scleroderma, 189

Paralysis, Infantile: See Poliomyelitis
neurologic complications during meningococcic meningitis treated with sulfonamide drugs, 201
Parasites: See under names of parasites and parasitic diseases
Parotitis; acute myocarditis in mumps (epidemic parotitis), 257
mumps, 183
Pellagra; powdered stomach in treatment of fatty liver and other manifestations of infantile pellagra; its significance with reference to problems of edema and steatorrhea in infants and in adults, 63
Penicillin, 114
bacteriologic studies of resistance to, 118
for various other diseases, 116
methods of administration, 117
sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161
Pericarditis, amebic, 88
Phillips, E.: Optimum dose of sulfadiazine in treatment of pneumococcic pneumonia, 22
Physicians, account of stock, 210
American College of, 398
Pinta, 187
Plague, pneumonic, 129
Plasmodium Vivax: See Malaria
Pleurisy; clinical syndrome in patients with pulmonary embolism, 234
Pneumococci: See also Pneumonia
sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161
Pneumonia, changes in central nervous system associated with encephalitis complicating pneumonia; clinical study, 146
clinical syndrome in patients with pulmonary embolism, 234
cold hemagglutination in primary atypical pneumonia and other common infections, 75
in association with malaria, 126
other forms of, 126
pneumococcic, 128
pneumococcic; optimum dose of sulfadiazine in treatment, 22
some observations on primary atypical pneumonia, 328
ulcerative tracheobronchitis following atypical pneumonia; report of cases, 93
viral pneumonia (viroid), 122
Pneumothorax in young adult males; descriptive statistics in 126 cases, 264
Poisons and Poisoning: See also under names of substances, as Benzene; Nitrotoluene; etc.
toxic effects of industrial poisons, 376
Poliomyelitis, 180
Porphyria: See Porphyrin and Porphyrin Compounds
Porphyrin and Porphyrin Compounds; acute porphyria; clinical and pathologic observations, 335
Power, M. H.: Effect of intravenously administered solution of acacia on animals, 39
Pregnancy, anemia associated with, 362
Price, A. H.: Streptomycin for certain systemic infections and effect on urinary and fecal flora, 269
Prisons and Prisoners; reversal of lingual atrophic changes with nicotinamide therapy, 31
Prothrombin: See Blood, coagulation
Psittacosis, 184
Pullen, R. L.: Tularemic meningitis; review of literature and report of case with postmortem observations, 163

Quinacrine: See Malaria

Rantz, L. A.: Etiology and pathogenesis of rheumatic fever, 131
Hemolytic streptococcus sore throat; detailed study of simultaneous infection of large number of men by single type, 278
Recruits: See Military Medicine
Reimann, H. A.: Infectious diseases; eleventh annual review of significant publications, 114, 178
Streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Respiratory Tract, diseases; control of air-borne infections, 122
diseases, experimentally produced, 129
diseases; infections of respiratory tract, 122
diseases, sulfonamide prophylaxis of, 120
diseases; treatment of acute infections of upper respiratory tract with sulfonamide compounds, 121
rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
study of water and heat loss from respiratory tract of man; methods: gravimetric method for measurement of rate of water loss; quantitative method for measurement of rate of heat loss, 308
Reticuloendothelial System: See Anemia; Jaundice; etc.
Rheumatic Fever, 182
etiology and pathogenesis of, 131
Rheumatism: See Arthritis; Rheumatic Fever
Rickettsia: See also Rocky Mountain Spotted Fever; Typhus
diseases, 185
Rocky Mountain Spotted Fever, 185
Rosenberg, D. H.: Acute myocarditis in mumps (epidemic parotitis), 257
Rosenblatt, P.: Healed subacute bacterial endocarditis, 1
Ross, G. T.: Studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
Rundles, R. W.: Blood; review of recent literature, 239, 358
Russi, S.: Small adenomas of adrenal cortex in hypertension and diabetes, 284
Ryon, J. B.: Congenital idiopathic methemoglobinemia; favorable response to ascorbic acid therapy, 299

Sagall, E. L.: Clinical syndrome in patients with pulmonary embolism, 234
Sayen, A.: Characteristic vascular pattern in patients with rheumatoid arthritis, 139
Schistosomiasis, 187
Schwartz, S. O.: Pernicious anemia in Chinese, 130
Scleroderma; esophageal lesions associated with acrosclerosis and scleroderma, 189
Sclerosis: See Arteriosclerosis
Scott, D. W., Jr.: Recurrent meningoencephalitis due to virus of lymphogranuloma venereum, 174
Scrub Typhus: See Tsutsugamushi Disease
Selzer, A.: Circulation in acute myocardial infarction, 54
Serum: See Blood
Sevringhaus, E. L.: Reversal of lingual atrophic changes with nicotinamide therapy, 31
Shigellosis: See Dysentery
Shock, clinical syndrome in patients with pulmonary embolism, 234
Sievers, R. F.: Congenital idiopathic methemoglobinemia; favorable response to ascorbic acid therapy, 299
Skin, blood supply; acute porphyria; clinical and pathologic observations, 335
Sleeping Sickness: See Encephalitis; Trypanosomiasis
Smalley, R. E.: Effect of intravenously administered solution of acacia on animals, 39
Smallpox, 184
Societies, Mississippi Valley Medical Society, 130, 256
Spingarn, C. L.: Cold hemagglutination in primary atypical pneumonia and other common infections, 75

Spink, W. W.: Etiology and pathogenesis of rheumatic fever, 131
Hemolytic streptococcus sore throat; detailed study of simultaneous infection of large number of men by single type, 278
Stats, D.: Putrid pulmonary abscess without foul sputum (shut-off pulmonary abscess), 154
Steatorrhea: See Feces, fat in
Stomach, achlorhydria in pernicious anemia, 242
Powdered: See under Liver
Stools: See Feces
Streptococci; etiology and pathogenesis of rheumatic fever, 131
healed subacute bacterial endocarditis, 1
hemolytic streptococcus sore throat; detailed study of simultaneous infection of large number of men by single type, 278
sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161
Streptomycin: See Actinomyces
Stuart, B. M.: Tularemic meningitis; review of literature and report of case with postmortem observations, 163
Sturgis, C. C.: Blood; review of recent literature, 239, 358
Sulfadiazine: See Pneumonia
Sulfonamides and penicillin; effect of combined therapy on experimental infections in mice, 161
Therapy: See Meningitis; Pneumonia; Respiratory Tract, diseases; Wounds; etc.
toxic effect of sulfonamide drugs including agranulocytosis, 380
Sultan, E. H.: Sulfonamide compounds and penicillin; effect of combined therapy on experimental infections in mice, 161
Suprarenals: See Adrenals
Syphilis: See also under names of organs and regions
paroxysmal hemoglobinuria due to cold hemolysin; observations with report of case of its occurrence in aerial gunner, 292

Teeth, extraction; sulfonamide prophylaxis in oral surgery, 121
Temperature: See Cold; Heat
Thalassemia: See Anemia, erythroblastic
Thiamine: See also Vitamins, B
relation of nutritional deficiency to cardiac dysfunction, 341
Thiouracil: See Thiourea
Thiourea, agranulocytosis due to thiouracil, 387
Thorax: See Heart; Lungs; etc.
Throat; hemolytic streptococcus sore throat; detailed study of simultaneous infection of large number of men by single type, 278
Thrombosis: See Embolism
Tongue, reversal of lingual atrophic changes with nicotinamide therapy, 31
Trachea; ulcerative tracheobronchitis following atypical pneumonia; report of cases, 93
Trinitrotoluene: See Nitrotoluene
Tropical Medicine; exotic diseases, 186
Trypanosomiasis, South American (Chagas' disease), 219
Tsutsugamushi Disease; absence of electrocardiographic changes in tsutsugamushi fever (scrub typhus); report of 200 consecutive cases, 217
Tuberculosis: See also under names of organs and regions, as Meninges; etc.
experimental, 129
pulmonary, 129
Tularemia, 185
tularemic meningitis; review of literature and report of case with postmortem observations, 163
Tumors: See Chorionepithelioma; and under names of organs and regions
Typhoid, 180
streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Typhus, 185

Ulcers: See also under names of organs and regions
sickle cell anemia in white patients with ulcers of ankles; report of 2 cases, 230
Undulant Fever; streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269
Urinary Tract: See also Kidneys
streptomycin for certain systemic infections and its effect on urinary and fecal flora, 269

Vaccinia: See Smallpox
Valley Fever: See Coccidioidomycosis
Variola: See Smallpox
Vasomotor System: See also Blood pressure
characteristic vascular pattern in patients with rheumatoid arthritis, 139
studies on hypertension; bioassay of vasoconstrictor substances in ultrafiltrates of citrated blood plasma from patients with normal blood pressures, patients with essential hypertension and patients made hypertensive by intravenous injections of angiotonin (hypertensin), 11
Veins: See Embolism; Vasomotor System; etc.
Venereal Diseases: See Syphilis; etc.
Venous Pressure: See Blood pressure
Vincent's Infection: See Fusospirochetosis
Viruses: See also Influenza; Lymphogranuloma Venereum; Pneumonia; etc.
other studies on, 184
Vital Statistics; account of stock, 210
Vitamins: See also Ascorbic Acid; Nicotinic Acid; Thiamine
B; possible relation of folic acid to agranulocytosis, 393
B_1: See Thiamine
C: See Ascorbic Acid
nutrition and anemia, 360
Vomiting; epidemic nausea, vomiting and diarrhea (viral or viroid dysentery?), 178

War: See Aviation and Aviators; Military Medicine; etc.
Water, rate of water and heat loss from respiratory tract of normal subjects in subtropical climate, 315
study of water and heat loss from respiratory tract of man; methods: gravimetric method for measurement of rate of water loss; quantitative method for measurement of rate of heat loss, 308
Weinroth, L. A.: Arteriosclerotic peripheral vascular disease in diabetes, 34
Winsor, T.: Habitus of patients with active sickle cell anemia of long duration, 47
Wintrobe, M. M.: Relation of nutritional deficiency to cardiac dysfunction, 341
Wolff, L.: Clinical syndrome in patients with pulmonary embolism, 234
Woofter, A. C.: Sickle cell anemia in white patients with ulcers of ankles; report of 2 cases, 230
Wounds, local therapy with sulfonamide compounds, 122

Yaws: See Frambesia